THE GROWTH OF THE BODY AND ORGANS IN ALBINO RATS FED WITH A LIPOID-FREE RATION

SHINKISHI HATAI

The Wistar Institute of Anatomy and Biology

Nearly seven years ago the writer attempted to raise stunted albino rats with the hope that a forced retardation of growth would induce some disturbance in the firm relation which normally exists between the weight of the body and of the central nervous system. The stunted rats were produced by feeding them with a minimum amount of nitrogenous food. It was found, however, that in this instance the artificial stunting did not modify the weight relation between the body and the central nervous system (Hatai '08). Although it was highly desirable to pursue this investigation further, yet on account of inconstancy and uncertainty of the outcome in raising stunted rats by the method employed, the investigation was postponed.

In 1911 Professors Osborne and Mendel published a series of remarkable papers in which the results of maintenance experiments by means of various isolated proteins were fully described. According to these investigators, albino rats about one-third grown can maintain their body weight for a considerable period without revealing any sign of nutritional or physical deterioration. This satisfactory and constant procedure for producing undersized rats renewed my interest in the problem mentioned.

During the past two years I have been so fortunate as to receive a number of stunted rats with their controls for examination. These came through the courtesy of Dr. McCollum, who raised the rats by feeding them with a 'lipoid-free ration.' These rats fall into two series: the series of 1913 and the series of 1914. The present paper contains the results of the anatomical examination of these interesting rats, and I take this

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opportunity to thank Dr. McCollum for his courtesy in putting these animals at my disposal.

The rats used were from those bred in the colony at The Wistar Institute in Philadelphia and sent to the University of Wisconsin. In each case rats belonging to the same litter were divided by Dr. McCollum into two lots with nearly identical body weights. The one lot was used for control and received the normal mixed ration, while the other lot, which was used for the experiment, received a specially prepared diet. As to the dietary formula, the following statements were kindly furnished by Dr. McCollum: The ration of the experimented rats which received the lipoid-free food was as follows:

Casein	18 per cent	Agar-agar	2 per cent
Lactose	20 per cent	Dextrin	56 per cent

The salts were as stated below:

Salt mixture	*Per 100 grams of ration*	
	No. 174 gm.	*No. 185* gm.
NaCl	0.808	0.168
$MgSO_4$ (anhydrous)	0.264	0.264
$NaH_2PO_4H_2O$	0.336	0.336
K_2HPO_4	0.936	0.964
$CaH_4(PO)_2H_2O$	0.528	0.528
Fe citrate	0.096	0.096
Ca lactate	2.000	1.300

The salt mixtures no. 174 and no. 185 were given at different periods in the case of both series.

At the end of the experiment these rats were shipped back to The Wistar Institute for the anatomical examination, where the writer determined the weights of the following organs: Brain and spinal cord, heart, lungs, kidneys, liver, spleen, alimentary tract, testes and ovaries, suprarenals, thymus, thyroid, hypophysis and eyeballs. Some of these organs were preserved for further histological examination. Besides the organs mentioned, the bones also were examined.

Although the methods employed in determining the relative amount of alteration in the various organs of the experimented rats, and also the technique for the preparation of the bones and separation of the encephalon into the four parts can be found

in my papers recently published (Hatai '13 and '14), I shall briefly restate the essential points.

The encephalon was divided into four parts in the following way:

1. *Olfactory bulbs.* The protruding portions of the olfactory tract with bulbs were cut from the rest of the encephalon by section of the tract just caudad to the bulb.

2. *Cerebrum.* The cerebrum is separated from the stem by a cut passing just in front of the dorsal edge of the anterior colliculi and just caudad to the corpus mammillare on the ventral surface.

3. *Cerebellum.* The cerebellum is separated by severing the peduncles.

4. *Stem.* The structure which is left after removal of these three parts mentioned above, is called the stem.

The bones were prepared as follows: The bones are freed from the main bulk of muscles and placed in a hot aqueous solution of 2 per cent 'gold dust washing powder.' After maceration for several hours at nearly 90°C., the remaining soft parts are removed. The bones thus prepared are gently wiped with blotting paper and are weighed. This gives the 'fresh weight.' These weighed bones are then dried at 95°C. for one week and the amount of moisture determined from the weight of the dried residue.

In order to determine the amount of modification following the experimental ration, we have employed our usual method of comparing the observed values with those found in a series of reference tables that have been compiled in this laboratory. These tables present for normal rats adequate data on all the organs and characters under consideration and in each case the graph representing the table can be expressed by a mathematical formula (Hatai '13; Hatai '14).

In making the comparison between the observed values and those in the tables—the body length is always used as the basal measurement and the weight of the body or organs as observed compared with the corresponding values given in the reference tables. In this manner comparison is made not only for the

experimented rats but also for those used as controls. The departures of the observed values from those in the tables having been observed in each case—the difference between that found for the experimented animals and that for the controls is obtained and this figure is used to indicate the amount by which the experimented animals have been modified.

Two examples will serve to illustrate this procedure. They are taken from table 3 — C, normal males, 1914 series: (1) On the 'mixed ration' the average tail length for the three rats is 172 mm., for the given body length, 196 mm. We expect from the reference tables a tail length of 165 mm. The observed value is therefore plus 4.2 per cent. The two rats on the "lipoid-free diet and egg fat" give a tail length of 151 mm. for a body length of 168 mm. From the reference tables we should expect a tail length of 139 mm. The observed value for the tail length of the experimented group is therefore plus 8.6 per cent. The difference between these two percentage shows the tail length in the experimented group to be 8.6 − 4.2, or 4.4 per cent greater than that of the controls. This is the value given in table 3. (2) Taking the brain weights for the groups just used we find by following the method employed above for the tail length, that the group on the "mixed ration" has a brain weight 4.8 per cent below the reference table value, while the group on the "lipoid-free ration plus egg-fat" has a brain weight which is 6.4 per cent deficient. Thus the brain weight in the experimented group is − 6.4 less − 4.8 or 1.6 per cent lower than in the controls. This is the value entered in table 2. All the percentage differences in the accompanying tables have been obtained in a manner similar to that illustrated by the two examples just given.

The only modification in procedure to which attention need be drawn is in the cases where the data from two series, 1913 and 1914, have been combined. In those cases the percentage deviation which is given in the table is the mean of the deviations for each series computed separately.

GROWTH OF BODY IN WEIGHT

The modifications of the growth of the body in weight due to the lipoid-free ration are shown in tables 1 and 2. Table 1 refers to the growth of the albino rats belonging to the 1913 series, while table 2 refers to the growth of the 1914 series. We note in both tables that the rats fed with the mixed ration made nearly normal growth in respect to their ages (see Donaldson '06). The spring

TABLE 1

Showing the weight of the body as modified by the lipoid-free ration compared with that of the rats raised on the mixed ration (1913 series)

DATE		MALES		FEMALES	
		Mixed ration (3)	Lipoid-free ration (4)	Mixed ration (4)	Lipoid-free ration (5)
1913					
April	16	94.7	93.8	85.0	76.2
	30	129.7	122.7	112.0	92.4
May	7	137.7	127.8	125.7	98.6
	14	153.7	134.8	146.7	108.4
	21	149.3	136.5	litters	107.4
	28	159.7	139.0	" "	108.4
June	4	166.3	140.7	" "	108.4
	11	173.3	131.5	129.0	103.8
	23	185.0	124.8	140.7	107.8
	30	196.7	134.2	143.3	109.0
July	7	209.7	139.7	151.0	111.6
	14	223.3	143.2	156.7	111.0
	21	222.7	149.2	161.0	107.2
	28	229.0	151.0	166.0	111.8
August	15	243.3	155.0	167.7	118.6
September	1	249.7	153.5	172.7	125.2

rats in 1913 made much better growth than the autumn rats in 1914. On the other hand, the experimented rats in both series made a noticeably poor growth when contrasted with the controls. In the 1913 series we notice that the experimented rats made continuous and steady growth throughout the period of experimentation, although the total amount of growth in weight was very slight. Curiously enough the experimented rats belonging to 1914 made a still smaller growth, and indeed in some cases the final body weight is no higher than the body weight at the begin-

TABLE 2

Showing the weight of the body as modified by the lipoid-free ration compared with that of the rats raised on the mixed ration (1914 series)

DATE	INTACT MALES			INTACT FEMALES			CASTRATED	
	Mixed ration (3)	Lipoid-free (2)	Lipoid-free and egg fat (2)	Mixed ration (3)	Lipoid-free (2)	Lipoid-free and egg fat (4)	Mixed ration (5)	Lipoid-free and egg fat (6)
November 26	96.3	79.0	90.0	82.3	95.0	80.0	84.2	94.8
December 3	118.0	100.5	105.5	101.7	102.0	82.0	100.8	103.2
10	133.3	104.0	105.0	113.7	98.0	84.5	115.4	105.7
17	147.0	106.5	107.0	121.7	103.5	85.2	122.4	108.2
26		106.5	118.0		107.0	90.2		108.5
January 1	171.3	104.0	119.5	136.3	111.0	91.5	150.4	111.2
7	181.0	103.0	121.0	143.0	103.5	92.8	162.2	110.0
14	182.3	102.0	111.0	144.3	107.0	91.2	167.6	106.5
February 3	198.7	96.5	113.0	141.0	111.5	95.2	169.6	108.2
16		87.0	108.0		105.5	93.3		104.8
24	207.3	84.0	107.0	166.3	108.0	93.2	179.0	111.5
March 5		84.5	110.5	litters	108.5	99.5		119.2
12	209.0	82.5	105.5	" "	105.0	96.5	182.6	117.7
19	212.0	82.5	105.5	" "	104.0	96.8	182.2	116.8
26	207.3	81.0	104.5	" "	103.0	93.0	184.6	115.2
31	210.3	79.0	102.5	155.7	105.0	94.5	187.6	119.3

ning of the experiment. This difference in growth in the two series may probably be due to the different physiological condition of the rats in these two series, combined with slight differences in the preparation of the ration. One point is clear, however: that the rats cannot continue the normal rate of growth on the lipoid-free ration in combination with the salt mixture which was used.

In table 2 we have also the data on the growth of the body of the albino rats which were fed first with the lipoid-free ration and later with the same ration to which a minute quantity of the egg-fat had been added. For convenience, these last mentioned rats will be designated simply as 'egg-fat series.'

It was found by McCollum and Davis ('13) that the rats whose body growth had ceased for a long period as the result of the lipoid-free ration, could be made to grow by the addition of a minute quantity of the extract of egg to the experimental ration. In order to see whether or not the rats thus treated would show any modifications other than those shown by the rats fed with the simple experimental ration, a small series was carried on. As will be seen from table 2, the 1914 rats given the extract of egg did not show the improvement in the growth of the body which was to be anticipated.[1] Thus the growth of the body is nearly identical in both the lipoid-free series and in the egg-fat series. Why in the present experiment the egg-fat series did not show a noticeable improvement in the growth of the body is not clear. However, from the fact that the control rats belonging to the 1914 series did not make satisfactory growth when contrasted with the 1913 series, we conclude that the failure to grow was

[1] "Our experience in feeding synthetic rations in this laboratory has convinced us that there exists a great variation in the vitality of individual rats as indicated by their ability to grow on such rations. It is unfortunate that practically all of the animals employed in the work here reported were not sufficiently vigorous to grow for a time in a nearly normal manner on the experimental ration, or to respond by a period of active growth when the ration was supplemented with egg yolk fats. We have individual rats in our colony at the present time which have been on the diet employed in the lipoid-free period with egg yolk fats added, during more than six hundred days, and which compare favorably with our stock rats in size and well-being."—E. V. McCollum.

probably due to a peculiarity of the rats rather than a peculiarity of the experimental ration.

Osborne and Mendel ('12) obtained normal growth of the rats with the ration from which the lipoid had been almost entirely removed. They carried the experiment for a considerable length of time by beginning with albino rats slightly over 30 days in age. In one series the experiment lasted for nearly 160 days. In every instance, so far as one can judge from the graphs, the body weight of the experimented rats was nearly identical with that of the control rats, while McCollum and Davis' rats, fed with the lipoid-free ration, did not grow at any period to the size of the controls (McCollum and Davis '13; see also present series).

This difference in growth between the rats of Osborne and Mendel, on the one hand, and those of McCollum and Davis on the other, was undoubtedly due to the nature of the inorganic salts and some extracts still contained in the food. The Osborne and Mendel rats received the inorganic salts from protein-free milk, while those of McCollum and Davis received the salts which were a laboratory mixture of pure chemicals. In reference to the varying effects of different salt mixtures McCollum and Davis state ('13) that

> "Young rats have been found to be very sensitive to variations in the character of the salt mixtures supplied, but with certain mixtures we have been able to obtain practically normal growth for periods varying from 70 to 120 days. Beyond that time little or no increase in body weight can be induced with such rations. The rats may remain in an apparently good nutritional condition on those rations for many weeks after growth ceases."

ANATOMICAL ANALYSIS

We now wish to present the results of the anatomical examination of these interesting rats reared by McCollum at the University of Wisconsin.

Although the growth rate was dissimilar in the two consecutive years, nevertheless it was found that the alterations shown by the various characters are nearly identical in the two series of experiments, and on account of this uniformity in the results, as well as to avoid unnecessary complication by presenting the

two series of data separately, I have combined the results. Consequently, unless otherwise stated, the figures given in the tables represent the averages of the two sets of data belonging to the 1913 and the 1914 series combined.

CENTRAL NERVOUS SYSTEM

If the lipoid-free ration is able to produce any alterations in the lipoid content of the organs, the central nervous system would naturally be expected to indicate such effects, since the central nervous system of the albino rat at about 200 days of age normally contains some 60 per cent of lipoid in the dried residue (Koch '13). This lipoid content is certainly greater in the nervous system than in any other organ (Koch '11). The weights of the central nervous systems of the experimented and of the control rats are shown in table 3 (see also page 16). As will be seen from this table, the weight of the brain with respect to the body length is generally slightly smaller in both the lipoid-free and egg-fat series. Only one exception is found in the female rats (B) fed with the lipoid-free ration in which the experimented rats show a slight over weight of 0.7 per cent. This slight increase is probably due to the abnormally small brain weight of the control rats, thus raising the relative weight of the central nervous system of the experimented rats. Indeed the normal brain weight of the female rats corresponding to the body length of 189 mm. should be 1.80 grams as against the observed weight of 1.73 grams, i.e., the observed weight of the control is nearly 4 per cent less than the normal brain weight. Without making any correction, however, we find on the average that the experimented rats show about 2 per cent less brain weight than the controls.

Similarly we find a reduction of 2.1 per cent in the weight of the spinal cord when compared with that of the control rats. This reduction of 2 per cent in weight in both the brain and spinal cord is somewhat greater than what we might expect from the normal fluctuation, nevertheless it is certainly far smaller than one might anticipate from the nature of the experiment. It seems reasonable, therefore, to conclude that the central nervous

TABLE 3

Showing the various measurements of the rats fed with the lipoid-free ration compared with the control rats

	LENGTH (MM.) OF		BODY WEIGHT (GRAMS)			WEIGHT OF		PER CENT WATER		SEX GLANDS	AGE	NO. OF RATS
	Body	Tail	Initial	Maxim.	Final	Brain	Sp. cord	Brain	Sp. cord		Days	
A. Normal males, 1913 and 1914 series combined												
Mixed ration	204	175.0	96	249	216	1.833	0.571	78.26	71.36	2.1820	193	6
Lipoid-free ration	179	158.0	84	133	118	1.678	0.466	78.04	70.78	0.8830	193	6
Per cent: Exp.-Control		4.2				−3.9	−1.4	−0.22	−0.58	−44.2		
B. Normal females, 1913 and 1914 series combined												
Mixed ration	189	164.0	84	165	151	1.730	0.528	78.28	71.79	0.0407	196	6
Lipoid-free ration	179	153.0	81	140	131	1.709	0.491	78.13	71.13	0.0303	195	8
Per cent: Exp.-Control		−0.7				0.70	0.30	−0.15	−0.66	−21.6		
C. Normal males, 1914 series												
Mixed ration	196	172.0	96	218	191	1.751	0.555	78.36	71.60	2.0320	186	3
Lipoid-free and egg fat	168	151.0	90	139	96	1.599	0.435	78.19	71.32	0.7830	186	2
Per cent: Exp.-Control		4.4				−1.6	−2.4	−0.17	−0.28	−44.2		
D. Normal females, 1914 series												
Mixed ration	185	161.0	84	157	137	1.729	0.529	78.21	71.82	0.0404	192	3
Lipoid-free and egg fat	162	147.0	79	128	100	1.569	0.416	77.98	71.23	0.0248	191	4
Per cent: Exp.-Control		5.1				−3.1	−4.9	−0.23	−0.59	−13.1		
E. Castrated, 1914 series												
Mixed ration (1)	193	168.0	84	188	182	1.780	0.548	78.25	71.50		189	5
Lipoid-free and egg fat (2)	175	158.0	95	119	131	1.709	0.480	77.98	71.13		188	6
Per cent: (2)–(1)		4.5				1.2	0.70	−0.27	−0.37			

system is adequately supplied with the necessary amount of the lipoids from the body and this fact in turn leads us to assume that the body has the ability to synthesize the lipoids from the non-lipoid materials. McCollum ('12) has demonstrated that the phosphorus needed by the animal for phosphatid formation can be drawn from inorganic phosphates, and that phosphatids can be synthesized anew in the animal body. Further investigation of McCollum ('12) indicates that certain complex lipoids of the lecithin type can be synthesized in large amounts by birds.

The percentage of water found in the central nervous system indicates also that the chemical composition has not been noticeably altered since the difference between the control and experimented rats is only 0.2 per cent in the brain and 0.5 per cent in the spinal cord; both in favor of the controls. It is to be noted, however, that a small reduction appears in all the experimented series, thus indicating a strong tendency to a slight modification.

To determine whether or not the reduction of 2 per cent in the weight of the central nervous system was mainly caused by the alteration in the white substance, in which the lipoids are predominant, the brain was divided into four parts and the weights and water content of those parts were found separately. The results of the investigation are shown in table 4. We notice from the table that although the percentage of water tends to be smaller in the experimented rats in all four parts, nevertheless the greatest relative reduction appears in the olfactory bulbs, the cerebellum comes next and the cerebrum and stem come in the order named. Thus the stem where the lipoid constituent is greatest is least modified and the olfactory bulbs and cerebellum, where the lipoid constituent is least, are most affected. From this we infer that the gray substance is most affected, and the white substance, in which one might anticipate the largest alteration, is least modified.

This fact of greater change of the gray matter is interesting, since it is found that during partial starvation with non-nitrogenous food for three weeks, the total brain weight shows a reduction of 5 per cent (Hatai '04) but the amount of myelin, as can be seen from the normality in the amount of alcohol and

TABLE 4

Showing the relative weights of the parts of encephalon and percentage of water found in them, of the rats fed with the lipoid-free ration, compared with the control rats

	INTACT MALE RATS			INTACT FEMALE RATS			CASTRATED RATS	
	Mixed ration	Lipoid-free ration	Lipoid-free and egg fat	Mixed ration	Lipoid-free ration	Lipoid-free and egg fat	Mixed ration	Lipoid-free and egg fat
Body length (mm.)	204	179	168	189	179	162	193	175
Weight of brain (grams)	1.833	1.678	1.654	1.730	1.707	1.570	1.780	1.709
Cerebrum	1.128	1.026	1.064	1.098	1.086	1.006	1.135	1.092
Stem	0.354	0.324	0.310	0.335	0.333	0.306	0.347	0.332
Cerebellum	0.260	0.226	0.230	0.244	0.239	0.215	0.250	0.239
Olfactory bulbs	0.053	0.041	0.051	0.052	0.040	0.042	0.048	0.046
Proportional weight of:								
Brain	100.000	100.000	100.000	100.000	100.000	100.000	100.000	100.000
Cerebrum	62.850	63.450	64.320	63.520	64.010	64.210	63.800	63.870
Stem	19.750	20.060	18.750	19.360	19.640	19.550	19.530	19.460
Cerebellum	14.440	13.980	13.890	14.130	13.990	13.650	14.040	14.000
Olfactory bulbs	2.970	2.510	3.050	2.990	2.350	2.600	2.630	2.670
Per cent of water:								
Brain	78.260	78.040	78.190	78.280	78.130	77.980	78.250	77.980
Cerebrum	78.950	78.750	79.040	78.950	78.940	78.720	78.980	78.870
Stem	74.500	74.340	74.390	74.480	74.530	74.510	74.560	74.410
Cerebellum	79.570	78.630	78.610	79.460	78.910	78.680	79.330	78.810
Olfactory bulbs	82.370	81.630	81.820	83.160	81.910	81.930	82.100	81.120
Age, days	193	193	186	196	195	191	189	188
Number of rats	6	6	2	6	8	4	5	6

ether extract as well as from the Weigert preparations, is not altered (Donaldson '11). We conclude therefore that the absence of lipoids from the ration does not affect the amount of the lipoids in the central nervous system, but on the contrary, the gray substance is affected. This alteration of the gray substance is similar to the effect of partial starvation on the brain of the albino rat.

SKELETAL SYSTEM

The skeletal system is naturally looked on as another structure which might show some alteration owing to the use of the artificial mixture of the pure chemicals contained in the ration in place of the salts normally present. For this purpose the following bones were examined: femur, tibia and fibula, humerus, radius and ulna. The results of the investigation are given in table 5. We note from this table that the ratio between body length and bone length and the ratio between body weight and bone weight are not altered. However, the water content found in these bones shows a distinct alteration in the experimented rats. The difference amounts to as much as 5.5 per cent in the case of the lipoid-free ration and 5.3 per cent in the case of the egg fat series; both in favor of the experimented rats. This difference of over 5 per cent is far greater than the usual incidental fluctuations. Furthermore, its constancy in direction in all these cases indicates that the chemical composition of the bones must be affected as the result of the experimental ration.

SEX GLANDS

The alterations thus far recorded are all of small magnitude, but we now come to the consideration of the one very obvious alteration. This is manifested by the testes and ovaries.

TESTES

We notice from table 3 that the experimented male has considerably smaller testes than the control. The difference amounts to as much as 44 per cent against the experimented. We notice also that the initial body weight of the experimented male rat

TABLE 5

Showing the weights and lengths of various bones of the rats fed with the lipoid-free ration compared with the control rats

	INTACT MALE RATS			INTACT FEMALE RATS			CASTRATED RATS	
	Mixed ration	Lipoid-free	Lipoid-free and egg fat	Mixed ration	Lipoid-free	Lipoid-free and egg fat	Mixed ration	Lipoid-free and egg fat
Body length, mm	196.000	169.000	168.000	185.000	178.000	162.000	193.000	175.000
Body weight, grams Obs.	191.000	95.000	96.000	137.000	128.000	100.000	182.000	141.000
Body weight, grams Calc.	181.000	113.000	111.000	161.000	142.000	106.000	172.000	125.000
Bone length	116.000	101.000	102.000	112.000	103.000	96.000	114.000	102.000
Bone weight Moist	2.688	1.660	1.905	2.340	1.868	1.569	2.564	2.007
Bone weight Dried	1.813	1.001	1.191	1.459	1.188	0.969	1.733	1.098
Per cent water, bones	32.560	39.730	37.500	34.870	36.420	38.220	32.440	45.290
Body length: Bone length	59.34%	59.70%	59.51%	60.27%	57.81%	59.26%	58.96%	58.46%
Body weight, calc.: Bone weight	1.48%	1.47%	1.72%	1.39%	1.32%	1.48%	1.49%	1.60%
Age, days	186	190	186	192	190	191	189	188
Number of rats	3	2	2	3	2	4	5	6

was 87 grams; this body weight calls for nearly 1.09 grams of testes, while the observed final weight is but 0.83 grams, thus showing a difference of nearly 23 per cent. We conclude therefore that the testes not only failed to grow during nearly six months of the special diet, but that there is a clear indication of an actual loss in weight.

OVARIES

In the case of the ovaries the difference between the controls and experimented is less than one-half of that found in the case of testes. The difference amounts to 17.4 per cent against the experimented. The initial body weight of the female rat was 80 grams; this body weight calls for nearly 0.015 grams of ovaries, while the observed final weight is 0.028 grams, thus showing an increment of more than 80 per cent during the experimental period of nearly six months. Thus it is clear that although the weight of the ovaries was 17 per cent smaller than that of the controls, nevertheless the ovaries of the experimented rats made steady growth, and indeed the final weight of the ovaries was nearly double the initial weight. The functional normality of the ovaries in the lipoid-free series is demonstrated by the fact that some of the females raised on the lipoid-free ration produced litters (McCollum and Davis '13).

EFFECT OF LIPOID-FREE RATION ON CASTRATED MALE RATS

The reduction of the testes in weight as the result of the experimental ration (1913 series) suggested that the same experimental ration when given to castrated male rats might produce a different alteration. To determine this point a series of castrated rats was sent to Dr. McCollum. Five of these castrated rats were fed with a mixed ration and six others were fed with the lipoid-free ration, to which latter a small amount of the egg-fat was added occasionally. The results of the investigation are given in tables 2, 3, 4 and 5. As is seen from table 2, the growth of the body in weight in castrates fed with the lipoid-free ration is similar to that of the intact rats fed in the same way. Thus castration, plus the lipoid-free ration, does not produce any other alterations, the testis excepted, than those shown by the intact rats fed in the same way.

We further note from tables 3 (E) and 4 that the central nervous system of the castrates fed with the experimental ration is not different from that of the intact rats fed in the same way Thus it is clear that the effect of the ration is not modified by castration. This conclusion applies also to the ratio between body length and bone length and the ratio between body weight and bone weight. The only difference is found in the percentage of water in bones of those castrates fed with lipoid-free ration.

We note that the difference in the water content of the bones between the castrates fed with the mixed ration and the castrates fed with the experimental ration, amounts to 13 per cent, which is much more than the difference between the intact rats on a mixed ration and those on the experimental ration. This greater difference of water content is found also in all individual cases. We conclude, therefore, that castration followed by the lipoid-free ration produces no further alteration than is found in the non-castrated rats fed with the same experimental ration, except in the water content in the bones. No explanation is possible for this singular result until further experiments have been made.

THE VISCERAL ORGANS AND DUCTLESS GLANDS

As has already been stated, the visceral organs and ductless glands, together with the eyeballs of these rats, were also examined. However, in view of the greater variability of these organs as well as the relatively small number of rats examined, I have decided not to attempt at this moment to interpret the alterations recorded by these organs. Nevertheless, for the reader who may wish to know the weight relations between the controls and the experimented rats shown by these organs, table 6 is given, where the results of the investigation are presented.

It may be important to add one word concerning the weight of the lungs. As will be seen from table 6, the weights of the lungs belonging to the experimented rats are always considerably smaller than those of the controls. This means that the lungs of the experimented rats were in a healthy condition and that the greater weights found in the controls were due not to sound lungs of larger size, but to a diseased condition. This fact must

be considered when interpreting the weight relations given by various other organs whose weights vary with the condition of the lungs (Hatai '13).

CONCLUSIONS

1. The lipoid-free ration diminishes the normal rate of the growth of the body.

2. The weight of the central nervous system shows a reduction of about 2 per cent as the result of the experimental ration. The percentage of water found in the central nervous system shows a very slight diminution.

3. The different parts of the encephalon are differently altered. In general, the parts where the gray substance is predominant are more affected than the parts where the white substance is predominant.

4. The weight and length of the longer bones with respect to body weight and body length are not modified. The percentage of water found in these bones, however, is constantly greater (5 per cent) in the experimented rats. This indicates that the chemical composition of the skeletal system has been somewhat altered.

5. The testes of the experimented rats showed not only a deficiency of 44 per cent as a result of six months of the lipoid-free diet, but there is a clear indication of actual loss in weight (23 per cent).

6. The ovaries of the experimented rats were smaller in weight by 17.4 per cent but no loss of the gland has occurred and growth was continuous.

7. The reactions shown by the lipoid-free ration and egg fat series are similar to those produced by partial starvation, especially with respect to the responses by the central nervous system and by the sex glands.

8. Although the lipoid-free ration causes a marked atrophy of the testes, yet in castrates on the lipoid-free ration no special alteration occurs which can be referred to castration, save the diminution of the solids in the bones.

9. Two incidental observations call for comment: (a) The loss of solids in the bones of the rats receiving a lipoid-free diet is of interest owing to the possible use of the phosphorus of the

TABLE 6

Showing the weights of visceral organs, ductless glands and eyeballs of the rats fed with the lipoid-free ration compared with the control rats

	BODY LENGTH	SUPRA-RENALS	THYROID	THYMUS	HYPOPHYSIS	HEART	KIDNEYS	LIVER	SPLEEN	ALIMEN. TRACT	LUNGS	EYEBALLS
	mm.	*grams*										
A. Normal males, 1913 and 1914 series combined												
Mixed ration	204	0.0332	0.0237	0.0934	0.0075	0.807	1.813	10.896	0.442	8.001	1.994	0.292
Lipoid-free ration	179	0.0271	0.0156	0.0483	0.0051	0.503	1.196	6.939	0.427	6.438	1.106	0.274
Per cent: Exp.-Control		8.0	−5.9	−26.7	−5.9	−12.9	−3.5	−10.4	32.0	−7.1	−38.4	−6.0
B. Normal females, 1913 and 1914 series combined												
Mixed ration	189	0.0489	0.0189	0.0700	0.0092	0.623	1.307	8.364	0.472	6.193	1.916	0.269
Lipoid-free ration	179	0.0338	0.0219	0.0761	0.0068	0.531	1.198	7.446	0.493	6.624	1.286	0.270
Per cent: Exp.-Control		−20.2	21.8	3.7	−6.3	10.8	5.9	2.1	24.1	14.2	−42.7	0.6
C. Normal males, 1914 series												
Mixed ration	196	0.0341	0.0217	0.0664	0.0069	0.783	1.727	8.786	0.425	7.160	2.184	0.283
Lipoid-free and egg fat	168	0.0261	0.0158	0.0610	0.0053	0.559	1.233	6.961	0.450	7.589	0.900	0.275
Per cent: Exp.-Control		4.4	4.5	−3.1	−10.4	4.1	9.3	12.9	58.8	36.8	−77.5	−2.7

TABLE 6—Continued

	BODY LENGTH	SUPRA-RENALS	THYROID	THYMUS	HYPOPHYSIS	HEART	KIDNEYS	LIVER	SPLEEN	ALIMEN. TRACT	LUNGS	EYE-BALLS
					D. Normal females, 1914 series							
	mm.	*gms.*										
Mixed ration.....	185	0.0468	0.0178	0.0485	0.0075	0.657	1.307	6.870	0.441	5.957	2.151	0.267
Lipoid-free and egg fat.....	162	0.0263	0.0134	0.0642	0.0054	0.478	1.242	5.454	0.352	5.804	0.726	0.253
Per cent: Exp.-Control.....		−5.3	2.1	9.0	11.2	−0.9	32.3	6.9	17.9	20.3	−115.6	−5.1
					E. Castrated rats, 1914 series							
Mixed ration (1)..	193	0.0357	0.0198	0.1108	0.0103	0.733	1.470	7.250	0.490	7.071	2.287	0.291
Lipoid-free and egg fat (2)....	175	0.0287	0.0157	0.0598	0.0073	0.557	1.327	7.072	0.441	7.020	1.116	0.282
Per cent: (2)-(1).		−2.6	−0.6	−23.8	−16.0	2.3	17.3	19.0	19.3	7.0	−81.2	−3.0

bone in the formation of lipoids. (b) On the lipoid-free diet, as well as in various forms of underfeeding, and after long-continued exercise, the rats become remarkably resistant to the lung infection which appears in the controls.

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THE SOURCE OF THE HYDROCHLORIC ACID FOUND IN THE STOMACH

FREDERICK S. HAMMETT

From the Departments of Anatomy and Biochemistry of the Harvard Medical School, Boston, Mass.

At the present time two opinions exist as to the source of the hydrochloric acid found in the stomach. The work of Miss Fitzgerald (1910) tends to show that the parietal cells of the gastric glands are the seat of the direct formation of the acid. Harvey and Bensley (1912), on the contrary, report that their experiments demonstrate that while these cells may form precursors of the acid, they do not produce the acid itself. Previous to these publications, Fränkel (1891) as well as Edinger (1880), by using dyes, had demonstrated an acid reaction below the surface epithelium of the gastric mucosa; but they could not accurately localize the source of the acid.

The disagreement between the conclusions of Miss Fitzgerald and of Harvey and Bensley appears to call for a critical review of their work, such as is here undertaken. In preparing it, I must thank both Dr. Frederic T. Lewis and Dr. Otto Folin for generous assistance.

Miss Fitzgerald found direct proof of the presence of acid in the lumina of the gastric glands, in the canaliculi of the parietal cells, and even in the parietal cells themselves. She found in these places a deposit of Prussian blue after injecting a ferrocyanide and a ferric salt into the ears of the animals experimented upon. This precipitate is formed in the presence of hydrochloric acid and the two salts mentioned. She found the precipitate in no other place than the immediate vicinity of the parietal cells. From her findings she draws the conclusion that the parietal cells of the gastric glands produce the hydrochloric acid found in the stomach.

Harvey and Bensley consider that Miss Fitzgerald's conclusions are unjustified, for reasons which are discussed seriatim in the following paragraphs.

1. The reaction is not constant. In some of Miss Fitzgerald's experiments the Prussian blue reaction was not obtained, and Harvey and Bensley, who repeated her work, likewise report unsuccessful experiments. No explanation, other than mere conjecture, is offered to account for these failures.

2. When present, the reaction is restricted to limited areas. Regional activity of the glands, decreased blood supply, and inhibition of secretory function due to the toxic effects of the injected salts, are all sufficient to explain this effect; the actual cause is unknown.

3. Within the areas which respond, the reaction occurs in only a few cells. The non-reacting cells can be considered either as resting cells, or as those which have already discharged their acid and are prevented from further activity by the toxic effect of the injected salts. It is to be noted that the parietal cells "of the deeper third of the gland tubules, that is, the third farthest from the free surface, never contained the Prussian blue." This may be correlated with the fact that the upper end or neck of the tubules is the source of new cells, as indicated by the presence of mitotic figures. And further, as stated by Kölliker (1902, p. 158), "the parietal cells are infrequent at the bottom of the glands, where they may be entirely absent; they increase in the body of the gland and are most frequent in the region of its neck." Thus the Prussian blue reaction appears to occur where the parietal cells are most active and most numerous.

4. The reaction occurs in other places than the gastric glands. Miss Fitzgerald did not find the Prussian blue precipitate in any tissues but those of the stomach wall. As a result of the toxic action of the injected salts a marked inflammation occurred. This would signify an increased permeability of the cell walls, and the secreted acid of the parietal cells could escape into the neighboring tissues as well as into the natural pathway. That

only traces of the precipitate were found outside of the glands proper is fairly convincing evidence of its intensive localization.

Although Harvey and Bensley found a precipitate in the liver, spleen, and blood vessels of the cardiac muscle, yet this precipitate may indeed not have been typical Prussian blue. We find, for example, that lactic acid, when added to a mixture of the salts in question, causes an atypical precipitate. This may explain Harvey and Bensley's apparently contradictory results of finding no precipitate in the heart's blood but finding it in the vessels of the heart muscle. In all probability it was precipitated there by the liberation of lactic acid from the dead muscle. Moreover we find that blood serum, blood plasma, and various salts which occur in the blood (sodium bicarbonate, sodium carbonate, sodium chloride, di-sodium phosphate, and mono-sodium phosphate, each in 0.1 per cent solution) may be added to a mixture of potassium ferrocynnide and iron and ammonium citrate without causing a precipitate. This is contrary to Harvey and Bensley's conclusion that "the Prussian blue is precipitated in the blood stream when solutions of these salts (sodium ferrocyanide and iron and ammonium citrate) are injected into it." At least there is no precipitate of Prussian blue when the salts are added to fresh normal blood in vitro.

Some of Harvey and Bensley's results with the Prussian blue reaction afford interesting evidence in support of Miss Fitzgerald's ideas. For instance, they find the Prussian blue precipitate on the mucous surface of the stomach, and prove that there is no backing up of the precipitate into the lumina of the glands; but occasionally they find the Prussian blue precipitate in these lumina. Therefore the Prussian blue must have been formed in the lumina of the glands. This necessitates acid, yet Harvey and Bensley deny the presence of acid in this situation. It seems as if they had furnished evidence of the presence of acid in the lumina of the glands of the gastric mucosa.

Furthermore, as might be expected on physiological grounds, they find that poisons and a decreased or restricted blood supply do not increase the formation of the Prussian blue precipitate. These influences would be expected to decrease the liability of

precipitate formation, so that only the favored locations would be able to respond. Both by their criticisms and their own work with the Prussian blue reaction, Harvey and Bensley appear to have strengthened greatly the conclusions of Miss Fitzgerald. After summarizing her experimental findings they state—"Our own results have confirmed these facts entirely." To prove that the cells of the gastric glands do not produce hydrochloric acid as such, Harvey and Bensley attempted injection experiments with various dyes; but this line of work did not yield satisfactory results

The next form of attack was to immerse portions of the gastric mucous membrane from a freshly killed animal in a solution of the dye *cyanamin* in normal sodium chloride solution. This dye yields distinctive colors for acid, akaline, and neutral solutions. With this method they found the contents of the gland cells to be alkaline. The acid reaction occurred on the surface of the mucosa and extended inward as far as the bottom of the gastric pits or foveolae. There it changed rapidly through neutral to alkaline, and so it extended through the lumina of the glands and into the secretory canaliculi of the parietal cells, which were thus strikingly demonstrated.

We have prepared cyanamin according to the directions given by Witt (1890) and have repeated Harvey and Bensley's experiment, obtaining similar results; but we draw different conclusions from these results.

Harvey and Bensley found that the concentration of the gland secretion is quite different from that of normal saline solutions. This being so, the laws of osmosis and diffusion come into play and we can not go on the supposition that the addition of the dye to the normal saline solution renders it isosmotic with the cell contents. Apparently no attempt was made to determine the relative concentrations of the reacting substances, e.g., dye and hydrochloric acid.

In order to determine for ourselves whether the dye or the hydrochloric acid diffused the more rapidly, experiments were conducted to that end. As might be expected, it was found that the acid diffused with by far the greater rapidity under conditions

approximating those in which the tissue was used. Having established this fact we have the following mechanical process as an explanation of Harvey and Bensley's results.

The hydrochloric acid secreted by the gland cells diffuses out of the cells, through the canaliculi and into the lumina towards the free surface, faster than the dye diffuses inward along the same path. Consequently the mucous surface of the tissue and the foveolar contents show acidity. The tissue, after removal from the organ, does not continue to perform its secretory function, nor does it excrete save by diffusion. This then leaves the cell contents alkaline, as is shown by the fact that the slowly moving dye stains the cells with the alkaline reaction. Supposing we have hydrogen weakly bound to protein and ionizing in the cell to $(H)^+$ and protein. We know we have sodium ions and chlorine ions present. $NaCl = (Na)^+ + (Cl)^-$. Removing the hydrogen ions and the chlorine ions we have an excess of sodium ions, thus making the cell contents alkaline.

The localization of the reaction between the dye and the acid is dependent upon the relative velocity of the participating constituents. Inasmuch as the acid has the higher velocity, we get the recorded results and a stable confirmation of Miss Fitzgerald's experiments and conclusions.

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STROPPING MACHINE FOR MICROTOME KNIVES

M. J. G.

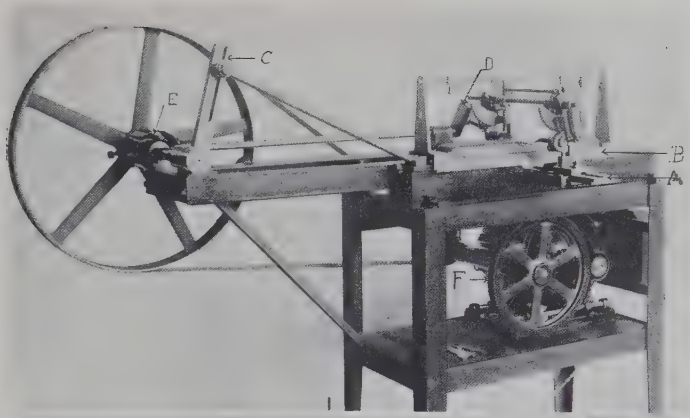
The Wistar Institute of Anatomy and Biology

This machine consists of two reciprocating carriers moving at right angles to each other, one (*A*) bearing the knife with the reversing mechanism and the other (*B*) carrying the strop. These carriers are actuated by cranks of unlike speeds (*C*, *D*) geared together (at *E*) and driven by a belt from a motor (*F*). The entire apparatus is constructed on an angle iron (2 inches × 2 inches) frame. Carrier *B* (fig. 2) is a heavy brass plate 19 inches long, $3\frac{3}{4}$ inches wide, and designed to carry a strop (*H*) 15 inches long and $2\frac{1}{4}$ inches wide. This carrier is grooved and accurately fitted to a base casting (*G*) upon which it travels. At each end of the carrier is a pillar (*I*). A $\frac{3}{8}$ inch steel rod (*J*) connects the tops of these pillars and carries a lug (*K*) at each end.

Carrier *A* is 12 inches long and is designed to carry a knife of any length from $3\frac{1}{2}$ inches to 7 inches. This carrier is mounted and moves in a base *L* secured at right angles to the base *G*.

O and *O* are vertical pillars carrying $\frac{1}{2}$ inch steel rods *M*, *M*, which move freely in a direction vertical to the plane of the strop. To the rods (*M*, *M*,) are attached by universal joints, the axis *Q* carrying the reversing mechanism *R* and the gear wheels *T*, *T*. Axis *S* (in which the knife forms the central portion) is also attached by universal joints to the vertical rods, *M*, *M*. It has also a gear wheel at each end meshing with the gear wheel *T* of the corresponding end of axis *Q*. The supporting pillar *O* may be adjusted upon carrier *A* to accomodate a longer or shorter knife. Axis *S* is made up of the knife *X* and the gear wheels *Y*, *Y*, with their hollow spindles as shown in figure 3. By means of pin *Z* the shank of the knife is prevented from turning in the hollow spindle of the gear wheel *Y*.

The gear wheel *Y* carries a wrist pin *XX* (fig. 3), to which is attached the lower end of the dash-pot *DP* (fig. 2); the upper member of the dash-pot is attached to axis *Q*. The dash-pot consists of an outer steel cylinder having an air outlet at the top (the size of which outlet may be changed by a screw) and an inner brass plunger with an air inlet at the bottom controlled by a valve. The springs on each side of the dash-pot tend to force the plunger into the cylinder by driving the air through the outlet at the top. The dash-pot springs, acting upon the wrist pin of the axis *S*, tend to tilt the knife edge downwards. The



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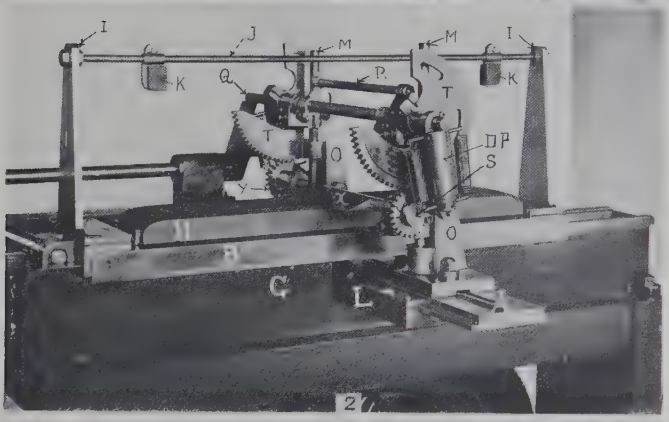

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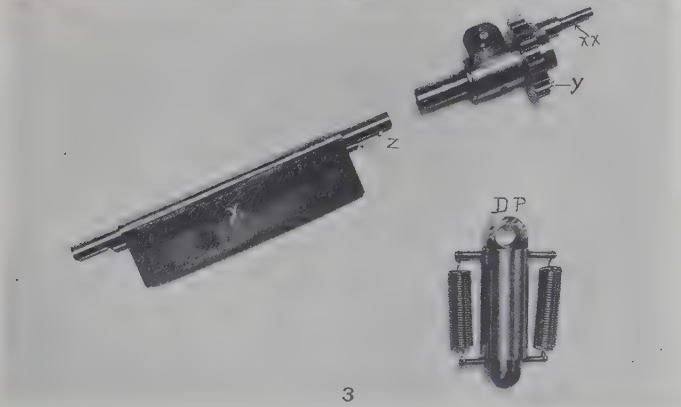

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dash-pot prevents the knife from striking too hard upon the strop when reversed.

The machine operates as follows: Carrier *B* moves to the left (fig. 2) while carrier *A* moves at right angles to carrier *B*. The knife is thus moved from end to end of the strop, and at the same time it moves part way across the strop. The gear ratio is such that the knife traverses the same path only once in every 137 strokes, thus bringing every portion of the knife edge in contact with every portion of strop surface. As carrier *B* passes to the end of its stroke to the left, the rod *R* comes in contact with lug *K*, and axis *Q*, with gear wheels *T*, *T*, is turned slightly anti-clock-wise. This movement gives the axis *S* a half turn clock-wise, thus throwing the knife edge to the right, at which instant the carrier *B* begins its stroke to the right. This process is repeated at each end of the stroke. The dash-pot springs pull the knife over firmly but slowly into its new position at each stroke. The knives used with this machine have a short shank at each end (fig. 3). The vertical rods *M*, *M*, move freely upward and downward carrying with them the knife and all the reversing mechanism. This permits the knife to follow over the surface of the strop and to conform to any irregularities which the strop may present.

The strop consists of a piece of Russia leather stretched over a piece of wood and secured at the ends. The strop is secured to the carrier by dowel pins and may be easily removed; the flesh side of the leather comes in contact with the knife. Any abrasive material may be smeared upon the strop, but experience proves that the best results are obtained by using castor oil in small quantities. The resulting edge is one free from the saw-toothed appearance and may be described as a polished line.

This machine was designed and constructed to do both honing and stropping. In actual use, however, it is found in most cases that a few moments on the honing stone is all that is necessary to prepare the knife for stropping. The practice followed at The Wistar Institute, where the machine has been in constant use for two years, is to give a knife about 10 or 15 minutes hand treatment on the honing stone, and then place it in the stropping machine for 10 to 30 hours.

Where any considerable amount of section cutting is done the time saved in sharpening microtome knives is a very considerable item. The machine was built at The Wistar Institute and may be duplicated by any good machinist. A quarter horse power motor running at 800 r.p.m. is used to drive the machine. The strop makes 25 complete strokes per minute.

A SIMPLE APPARATUS FOR MICROSCOPIC AND MACROSCOPIC PHOTOGRAPHY

DANIEL DAVIS

From the Physiological Laboratory, Johns Hopkins University

THREE FIGURES

One of the pioneers in photomicrography was Leon Foucoult,[1] who in 1844 was the first to make daguerreotypes with the microscope by means of electric light. Since that time many forms of photomicrographic instruments have been designed. All of these cameras, however, can be divided into three types, the horizontal, the vertical, and the convertible, each type possessing advantages peculiar to itself.

Of these three classes of apparatus the horizontal is certainly the oldest and perhaps still the most popular. Its chief advantage is that it allows unlimited bellows extension. This was at first of primary consideration, for in the early days of photomicrography the water and the oil immersion objectives, to say nothing of the apochromat, were unknown. Consequently, the only way of obtaining high magnifications was by means of a comparatively low-power objective used without any ocular, the image being projected a considerable distance onto the plate. This method is still of value when great depth of field is desired. Furthermore, it is somewhat easier to obtain critical illumination with the horizontal camera, since the beam of light can be projected directly into the microscope, obviating the use of the mirror. A simple and efficient camera of this type has been constructed by Parker,[2] while very complete and ingenious machines have been described by Barnard[3] and Buxton.[4]

Owing to its greater compactness the vertical camera is often preferred. With the modern highly perfected lenses great bellows extension is no longer essential for high-power work. This is due to the fact that the image formed by an apochromatic objective can be very greatly enlarged by means of a compensating or projection ocular without losing any of its original sharpness. The vertical camera is of course the only type to be used in photographing specimens in liquid. Perhaps the best camera of this type is the Van Heurck, but a very simple

[1] E. J. Spitta: Jour. Quekett Micr. Club, London, 1907–08, n. s. x, 51.

[2] H. B. Parker: Bulletin No. 7, of the Hygienic Laboratory, Washington. 1902, p. 7.

[3] J. E. Barnard: Tr. Jenner Inst. Prevent. Med., London, 1899, 2 s, p. 248.

[4] B. H. Buxton: Jour. Applied Micr., Rochester, 1901, vol. 4, p. 1366.

outfit has been described by Borden,[5] while Terras[6] has designed a vertical camera resting on the floor in which the microscope is carried on a very low shelf, thus making possible the convenient use of long bellows extensions. The simplest upright camera is a light box fitting directly on the tube of the microscope. The description of an aluminum camera of this character has recently been given by Wilson.[7]

It is to possess the advantages peculiar to each of the above types that the numerous forms of convertible cameras have been designed, and it is to this type that the machine which I wish to describe belongs. It is obvious, however, that such an instrument can not also possess the many little refinements peculiar to either the horizontal or the vertical form. This machine is the result of three years' experimentation. If it merits a description it is because it is so easily and so cheaply built, and because it is so simple and accurate of manipulation.

The stand consists of two parts, the optical bench, and the camera rest. Each is of the same width, length, and thickness. The optical bench is formed from two pieces of $\frac{7}{8}$ inch poplar 3 feet long and 3 inches wide. These strips are screwed and glued to two cross-pieces, one at each end, each piece measuring $\frac{7}{8}$ x 3 x $8\frac{1}{4}$ inches. There are two other cross-pieces only 2 inches wide, equally spaced across the bottom. In order to make the bench still more rigid, a batten measuring 3 feet x $\frac{7}{8}$ x $1\frac{3}{4}$ inches is fastened along either side. These prevent the bench from sagging. On the top of this bench is a track on which the various auxiliary condensers slide. This track is formed of two similar brass tubes, 3 feet long, $\frac{5}{8}$ inch outside diameter, and $\frac{1}{16}$ inch thick. The distance between the centers of these tubes is $5\frac{1}{4}$ inches.

The camera rest is formed of four poplar strips, all of which are 3 feet long and $\frac{7}{8}$ inch thick. Two of the strips are 1 inch while the others are 2 inches in width. Only two cross-pieces are used, one at each end. These supports are of exactly the same dimensions as the end cross-pieces fitted to the optical bench. Battens are similarly fastened to the sides, the 1 inch strips coming next to them, the 2 inch strips then being fastened in place with a space of 1 inch between them and the narrower pieces. This will leave just enough room between the center pieces for the bolts holding the camera on the rest, while the tubes on the optical bench fit into the spaces on either side when the stand is folded up. The stand complete is shown in figure 3. It will be seen that the parts are fastened together with bracket hinges, while the camera rest can be clamped in a vertical position by means of oak side braces held in place by small bolts fitted with thumb nuts. The exact dimensions and position of these braces are immaterial; depending somewhat on the size and type of camera used, but the pieces should be slotted at one end as shown, so that to lower the rest to the horizontal position it will be necessary only to loosen the thumb nuts.

[5] W. C. Borden: Am. Month. Micr. Jour., Washington, 1896, vol. 17, p. 193.

[6] J. A. Terras: Proc. Scot. Micr. Soc., London and Edinburgh, 1899–1903, vol. 3, p. 210.

[7] L. B. Wilson: American photography, Boston, 1914, vol. 8, p. 204.

Furthermore, it is important to arrange the braces so that the camera may be clamped to either side of the rest. This rest will accommodate nearly any style of plate camera not larger than the $6\frac{1}{2} \times 8\frac{1}{2}$ inch size with a bellows extension of not more than 3 feet.

The lamp and the various auxiliary condensers are supported on the track of the optical bench by means of geometric slides, as described by Barnard.[8] The slide is made from a piece of $\frac{7}{8}$ inch poplar $3\frac{1}{2}$ inches wide and $10\frac{1}{2}$ inches long. Four inches from one end across the bottom of this piece is cut a V-shaped groove $\frac{1}{2}$ inch deep. The sides of this groove are at 90° to each other; one of the tubes of the track fits into this groove, and it is by this means that the slide is kept in alignment.

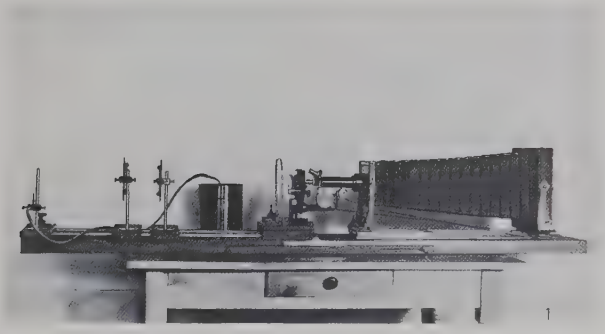


Fig. 1 Camera in horizontal position. The U-shaped frame on the microscope table is used to support the ray filter. The condensing lenses, a plano-convex and the meniscus, are arranged for high-power work.

The other end of the support resting on the other tube is cut away till the slide sits level upon the track. There is, of course, one such support for each lens that is to be carried on the optical bench, the lens being attached to an upright rod of convenient length fastened to the slide, as shown in figure 2. I have found it convenient to use three lenses, all 4 inches in diameter. Two are plano-convex of 12 inches focal length, while the other is a double convex lens of 18 inches focal length, thus forming a simple Kohler condensing system, as suggested by Barnard.[8] These lenses are mounted on $\frac{1}{8}$ inch sheet iron rings $5\frac{1}{2}$ inches in diameter with a $3\frac{3}{4}$ inch hole in the center, the lens being supported by three equally spaced machine screws fitted with washers and short spiral spring sleeves. A slotted 6 inch iron rod of the same diameter as the upright on the slide (in this case $\frac{3}{8}$ inch) is riveted radially to the ring, this rod being fastened to the upright by means of an adjustable clamp.

[8] J. E. Barnard: Practical photomicrography, Edward Arnold, London, 1911.

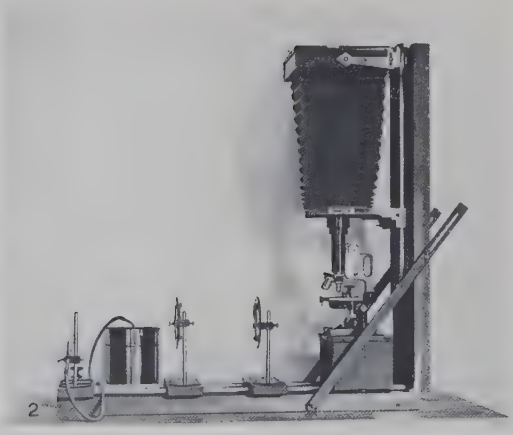


Fig. 2 Camera in vertical position. The two plano-convex condensing lenses arranged for medium-power work. For low-power work the meniscus lens is substituted for the plano-convex lens nearer the light.

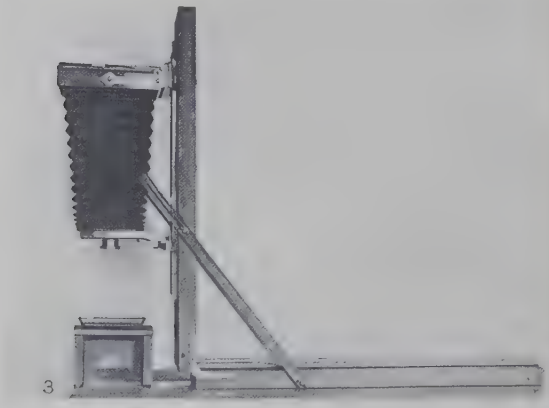


Fig. 3 Camera fitted with a wide angle lens and fastened to the back of the rest for the photographing of embryos.

Acetylene has proved an excellent illuminant. The light is very actinic, and perfectly steady. A ½ foot burner is ample, and the gas can readily be made in any simple generator. The gas should be passed through a fairly large bottle before being fed to the burner. This serves to maintain a steady pressure as well as cooling the acetylene and allowing the condensation of water. Such a light used with the condensers just described has proved ample for magnifications of over 1000 diameters.

The microscope is supported on a table, the legs of which just straddle the track on the optical bench to which this table is clamped. Having been properly centered, the microscope is fastened in position by means of an oak strip extending across the horseshoe base. This strip is bolted to the microscope table. Two small blocks are attached to the table, as shown in figure 1. These fit snugly against the side of the horseshoe base, serving as guides to the correct position of the microscope should it be removed from the stand. All fastenings used on this table should of course be fitted with thumb nuts. The height of the table must be such as to cause the optical axes of the microscope and of the camera to coincide. The condensing lenses and burner are then adjusted. When the camera is placed vertically another table is used of such a height as to make the optical axis of the condensing system center upon the microscope mirror. The condensers thus need no readjustment when changing the camera from the horizontal to the vertical position.

In use this outfit has proved very satisfactory. When in the horizontal position the field can be examined by sliding back the camera front, or perhaps more conveniently by removing the microscope from the stand. By means of the guide blocks the microscope can readily be replaced in correct alignment. When using the greatest bellows extension, focusing is accomplished by means of a waxed silk cord passing around a little pulley fitted to the knob of the fine adjustment. When used vertically the microscope, once having been adjusted, is not moved. If it is desired to examine the field it is but necessary to raise the camera front a few inches and then lower the camera rest out of the way into the horizontal position. When ready to photograph the camera can quickly be swung up over the microscope. Finally, by clamping the camera to the back of the stand, as shown in figure 3, and using a photographic lens of short focal length, a most convenient arrangement is obtained for copying drawings, or photographing embryos or other macroscopic specimens.

INCREASE IN PRICE OF JOURNALS

In order to extend and improve the journals published by The Wistar Institute, a Finance Committee, consisting of editors representing each journal, was appointed on December 30th, 1913, to consider the methods of accomplishing this object. The sudden outbreak of European misfortunes interfered seriously with the plans of this committee. It was finally decided, at a meeting held December 28th, 1914, in St. Louis, Mo., that for the present an increase in the price of these periodicals would not be unfavorably received, and that this increase would meet the needs of the journals until some more favorable provision could be made.

This increase brings the price of these journals up to an amount more nearly equal to the cost of similar European publications and is in no sense an excessive charge.

The journals affected are as follows:

THE AMERICAN JOURNAL OF ANATOMY, beginning with Vol. 18, price per volume, $7.50; foreign, $8.00.

THE ANATOMICAL RECORD, beginning with Vol. 9, price per volume, $5.00; foreign, $5.50.

THE JOURNAL OF COMPARATIVE NEUROLOGY, beginning with Vol. 25, price per volume, $7.50; foreign, $8.00.

THE WISTAR INSTITUTE OF ANATOMY AND BIOLOGY

36TH STREET AND WOODLAND AVENUE

PHILADELPHIA, PA.

PROCEEDINGS OF THE AMERICAN ASSOCIATION OF ANATOMISTS

THIRTY-FIRST SESSION

At the Washington University Medical School, St. Louis, Mo., December 28, 29 and 30, 1914

Monday, December 28, 9.30 a.m.

The thirty-first session of the American Association of Anatomists was called to order by President G. Carl Huber, who appointed the following committees:

Committee on Nominations: G. S. Huntington, chairman; Irving Hardesty, Florence R. Sabin.

Auditing Committee: T. G. Lee, chairman; A. T. Kerr.

President G. Carl Huber, in recognition of the great loss the Association had sustained in the death of Professor Minot, suggested that some arrangement be made for expression from the Association. Prof. F. T. Lewis moved that the following committee be appointed to draw up appropriate resolutions: Chairman, Prof. George S. Huntington; Members, Professors G. Carl Huber and R. J. Terry, the resolutions to be presented at a future meeting of the Society.

Tuesday, December 29, 12.00 m. Association business meeting, President G. Carl Huber, presiding.

The Secretary reported that the minutes of the Thirtieth Session were printed in full in The Anatomical Record, volume 8, number 2, pages 69 to 145, and asked whether the Association desired to have the minutes read as printed. On motion, seconded and carried, the minutes of the Thirtieth Session were approved by the Association as printed in The Anatomical Record.

Prof. T. G. Lee reported for the Auditing Committee as follows: The undersigned Auditing Committee has examined the accounts

of Dr. Charles R. Stockard, Secretary-Treasurer of the American Association of Anatomists, and finds the same to be correct, with proper vouchers for expenditures, and bank balance on December 23 of $285.85. (Signed) T. G. Lee, A. T. Kerr; St. Louis, Mo., December 29, 1914.

The Treasurer made the following report for the year 1914:

Account of G. Carl Huber, former Secretary-Treasurer, closed January, 10, 1914

Balance on hand December 26, 1913, when accounts were last audited	$213.03	
Collections made from December 26 to January 10, 1914	55.00	
Total deposit	$268.03	$268.03
Expenses of Secretary-Treasurer attending Philadelphia Meeting, December 29–31, 1913	$49.00	49.00
Balance sent by draft to Charles R. Stockard, Secretary-Treasurer		$219.03
Amount of draft	$219.03	
Receipts for dues, 1914	1520.20	
Total deposits for 1914	$1739.23	$1739.23
Expenditures for 1914:		
Postage ($45.42), printing ($34.00)	$79.42	
To 305 subscriptions to 1 volume of the American Journal Anatomy and 1 volume of the Anatomical Record @ $4.50	1372.50	
To collections and exchange on foreign and domestic drafts	1.46	
Total expenditures	$1453.38	$1453.38
Balance		$285.85

Balance on hand, deposited in the name of the American Association of Anatomists in the Corn Exchange Bank, New York City, December 23, 1914.

On motion the reports of the Auditing Committee and the Treasurer were accepted and adopted.

The Committee on Nominations, through its Chairman, Prof. George S. Huntington placed before the Association the following names for members on the Executive Committee for terms expiring in 1918: J. L. Bremer and H. von W. Schulte.

On motion the Secretary was instructed to cast a ballot for the election of the above named officers.

The Secretary presented the following names recommended by the Executive Committee for election to membership in the American Association of Anatomists:

Wayne Jason Atwell, A.B., Instructor in Histology, 1335 Geddes Avenue, Ann Arbor, Michigan.

Henry K. Davis, A.B., A.M., Instructor in Anatomy, *Cornell University Medical College, Ithaca, New York.*

Arnold Henry Eggerth, Assistant in Histology, *University of Michigan, Ann Arbor, Michigan.*

J. F. Gudernatsch, Ph.D., Instructor in Anatomy, *Cornell University Medical College, New York City.*

Elmer R. Hoskins, A.B., A.M., Instructor in Anatomy, *University of Minnesota, Minneapolis, Minnesota.*

Charles Eugene Johnson, Ph.D., Instructor in Comparative Anatomy, *Department of Animal Biology, University of Minnesota, Minneapolis, Minnesota.*

Beverly Waugh Kunkel, Ph.B., Ph.D., Professor of Zoology, *Beloit College, Beloit, Wisconsin.*

Paul Eugene Lineback, A.B., M.D., Teaching Fellow in Histology and Embryology, *Harvard Medical School, Boston, Massachusetts.*

C. C. Macklin, M.D., Assistant in Anatomy, *Johns Hopkins Medical School, Baltimore, Maryland.*

William Eli McCormack, M.D., Instructor in Embryology and Histology, *University of Louisville, Louisville, Kentucky.*

D. Gregg Metheny, M.D., *Jefferson Medical College, Philadelphia, Pa.*

Roy L. Moodie, A.B., Ph.D., Assistant Professor of Anatomy, *University of Illinois, Chicago, Illinois.*

Henry R. Muller, M.D., Assistant in Anatomy, *Johns Hopkins Medical School, Baltimore, Maryland.*

Jay A. Myers, A.B., Ph.D., Instructor in Anatomy, *University of Minnesota, Minneapolis, Minnesota.*

H. W. Norris, B.S., A.M., Professor of Zoology, *Grinnell College, Grinnell, Iowa.*

James Wenceslas Papez, B.A., M.D., Professor of Anatomy, *Atlanta Medical College, Atlanta, Georgia.*

Franklin P. Reagan, A.B., *Princeton University, Princeton, New Jersey.*

Randolph Tucker Shields, A.B., M.D., Dean, *University of Nanking, Medical School, Nanking, China.*

P. A. West, B.A., *Johns Hopkins Medical School, Baltimore, Maryland.*

Harry Oscar White, M.D., Professor of Anatomy, Histology and Embryology, *Medical Department, University of Southern California, Los Angeles, California.*

John Locke Worcester, M.D., Instructor in Anatomy, *University of Michigan, 1214 Willard, Ann Arbor, Michigan.*

On motion the Secretary was instructed to cast a ballot for the election of all the candidates proposed by the Executive Committee. Carried.

The following proposed amendment, having been a matter of record at the last meeting, was presented for action by the Association. "These officers shall be elected by a ballot at the annual meeting of the Association and their official term shall commence with the close of the annual meeting."

"At the annual meeting next preceding an election, the President shall name a nominating committee of three members. This Committee shall make its nominations to the Secretary not less than two months before the annual meeting at which the election is to take place. It shall be the duty of the Secretary to mail the list to all members of the Association at least one month before the annual meeting. Additional names for any office may be made in writing to the Secretary by any five members at any time previous to balloting."

The Association voted the adoption of this amendment.

The following proposed amendment, also recorded at the last meeting, was presented for action by the Association: Amendment of Article VI of the Constitution. The first sentence of the article "the annual dues shall be $5.00," it is proposed to amend to read "the annual dues shall be $7.00."

The Association voted the adoption of this amendment.

It was pointed out by Dr. M. J. Greenman, Director of The Wistar Institute, that since the dues of the Anatomists were now advanced to $7.00 per year, the members of the Association would receive all numbers of The American Journal of Anatomy and The Anatomical Record, six numbers of the Journal of Anatomy and twelve numbers of the Record yearly.

The special Committee on Pre-Medical Work in Biology presented through its chairman, Dr. H. McE. Knower, the following report:

Your Committee was appointed to confer with the Zoölogists to ascertain what coöperation may be expected toward standardizing work in Biology required of students looking forward to the study of medicine; and to formulate the considerations which would seem practical to incorporate in plans for such courses.

The Zoölogical Society promptly appointed a committee for this conference, and the following questions were discussed, not only with this committee, but with a number of other representative members of the Zoölogical Society. Besides this, published statements of courses and of discussions on this subject were examined.

The following questions seemed to be most important:

Question 1. Is the work given in different colleges in the elementary, general course in Biology adapted to satisfy the requirements of premedical training in this subject?

Question 2. Is it possible to so select and standardize the work of the first year in Biology in different colleges as to make it uniform, and to include, here, all needed to make it *an adequate* course?

Question 3. If an ideal course, including sufficient preliminary work can not be secured within the one year period advocated, what principles should be urged to govern the planning of the biological work of students looking forward to the study of medicine, so that they will profit most by the training of the first year, and be best prepared to follow this up in special departments of Biology more directly related to medicine.

Question 4. What additional work is to be advised, which is not to be obtained in the first year's general course?

Both committees agree that it is of the first importance to urge the selection of only thoroughly trained scientific men as teachers for this work. Such men can be trusted to insist on real scientific methods and to select the best material and treatment to give the beginner a practical introduction and basis for further work.

Beyond this point, however, the committees were unable to proceed. The Zoölogists suggested that the Anatomists should draw up a statement of what they desire the Zoölogists to do, in preparing students for anatomy. After this has been done, the Zoölogists are ready to consider how far it is practical to meet these needs. Several attempts have been made in this direction, and your committee submits the following statement to the Association for its approval and transmission to the Zoölogists.

At the present time a one-year's course in biology is generally required as a preparation for the work of the medical school. This study of biology must serve as a preparation for medical work in physiology, pathology, bacteriology and parasitology, as well as anatomy, and it may fairly be questioned whether a single college course is adequate for this purpose. The study of botany alone is obviously insufficient, and the domain of zoölogy is so vast that much care should be exercised in the choice of the phases of the science to be presented to young students. Courses which are primarily experimental and deal with the functions and reactions of animals, although excellent in preparation for the physiological work of the medical school, are not the proper basis for the study of human anatomy. It is the purpose of this report to point out only those features of the college preparation which experience has shown to be desirable, and in fact essential, for the successful study of gross and microscopic anatomy.

No uniform or stereotyped preparatory course is recommended, for it is recognized that every teacher should give special attention to those subjects and groups in which he is particularly interested, and to the knowledge of which he has contributed by his own researches. Success depends in large part upon the ability of the teacher, but the following purposes of instruction should not be forgotten if the preparatory work is to satisfy the requirements of anatomy.

1. Students frequently begin the study of human anatomy with an insufficient knowledge of the lower forms of animal life. The broad knowledge of the various classes of animals and of invertebrate and lower-vertebrate morphology, which was the inspiration of the great anatomists of the past, is now too often replaced by vague considerations of the method of science and ideals of observation. A return to the study of animals, as objects of interest in themselves, apart from theoretical considerations and possible relations to human society, is therefore recommended. The student should obtain a synoptic knowledge of the animal kingdom, and should be able to classify in a general way, and to describe the life histories of the common forms of animals, aquatic and terrestrial, which may be collected in his locality. A beginning in such work may well be made by the student independently, or perhaps in high-school courses, but such fragmentary and elementary studies should be supplemented by a thorough college course. The first-hand familiarity with animals thus obtained should serve as the basis for all further work.

2. As a result of the knowledge of genera and species which the student should have obtained directly for himself, by studying some group of animals or plants; questions of the origin of species and of the relation of the great classes of animals to one another are inevitably before him as philosophical problems. Collateral reading then becomes as necessary for the biologist as for the man of learning in any other branch of knowledge. Selected works of Lamarck, Darwin, Huxley, Mendel, and others should be freely consulted. This literature, which in its influence upon human thought has far outspread the bounds of biology, should not be nelgected by the student of zoölogy, whose particular heritage it is. Since the idea that science cannot be read, and that there is no knowledge in books, is often taught as a cardinal principle, it has come about that students of zoölogy have little knowledge of, or respect for, the writings of the makers of their science.

3. Before beginning the study of human histology, every student may reasonably be expected to be familiar with the use of the microscope and with the simpler methods of preparing specimens for microscopic examination. This technique can be learned in connection with various courses, perhaps the most useful of which is a general study of the cell with a comparative study of the elementary tissues. The maturation of the germ cells and the processes of fertilization and segmentation cannot be properly presented in the medical curriculum, and these fundamental biological phenomena should therefore be observed in college courses. The development of the chick, which was studied primarily by physicians to explain the growth of the human embryo, can likewise receive little attention in the medical school. These subjects are all very desirable in themselves, and if studied by laboratory methods, will supply the requisite skill in the use of the microscope.

4. In preparing for human dissection, comparative anatomy should be studied with the same standards of thoroughness which obtain in

the dissecting room. The student should learn to dissect rapidly and well, and to record with careful drawings and brief descriptions the forms and relations of the structures which he has disclosed. But such studies are not useful merely for their methods. A knowledge of comparative anatomy, including especially the anatomy of the lower vertebrates, is indispensable for understanding the structure of the human body. For other reasons also, human anatomy must be treated as an advanced study. The State does not provide bodies for dissection in order that untrained students may learn from them those elementary facts, which may be understood equally well by dissecting cats or rabbits. "It is absurd," says President Eliot, "to begin with the human body the practice of dissection." And the value of dissection is so great in relation both to medicine and surgery, that an adequate preparation should be required. For the study of anatomy, in the words of Lord Macaulay, "is not a mere question of science; it is not the unprofitable exercise of an ingenious mind; it is a question between health and sickness, between ease and torment, between life and death."

5. Finally, these recommendations may be summarized as a plea for a more thorough study of zoölogy on the part of those planning to enter the medical schools. The zoölogical courses should not be abridged and popularized in order that time may be saved for other pursuits, or that the science may seem more attractive to college youth. Courses in anatomy and physiology which duplicate the work of the medical school, and courses in "medical zoölogy," ought not to be substituted for the strictly zoölogical university courses. The science of zoölogy is of such great service to students of medicine that it deserves a large place in their undergraduate studies. With medical anatomy, it constitutes "a subject essentially one and indivisible;" and the penalty for its neglect is inadequate preparation for medical practice.

St. Louis, Missouri Committee: H. McE. Knower, *Chairman*
December 29, 1914 F. T. Lewis
W. H. Lewis

In the following summary, the Chairman of the Committee has rearranged the main points of the above report in groups, to correspond to the four questions proposed at the beginning; so that a more definite idea may be secured of the manner in which these are answered. In assembling the answers to the different questions the exact sense of the report itself has been retained. In answering questions 3 and 4 an effort has been made to indicate what we may reasonably expect to include in the first year, and what should be advised in addition.

1 and 2. The first two questions formulated by the Committee are answered in the negative; that is, a one-year's course is not regarded as sufficient, and a uniform, standardized course seems undesirable. An introduction to the subject through special courses in selected "medical zoölogy" is also disapproved.

3. (a). In regard to the third question, it has seemed necessary to urge a more thorough knowledge of the morphology of lower forms of

animals and their life histories. While the Anatomists in adopting this statement as given in the report, undoubtedly, expect the physiological aspects of these mechanisms to be considered as necessary accompaniments of such first hand familiarity with animals; it is urged in the report that the introductory college course shall not be "primarily physiological." It is earnestly desired that the work should involve a rigorous grounding in comparative morphology, especially of lower forms, which furnishes not only the best basis for human anatomy, but is a very essential preliminary for comparative and human physiology. (b). It is urged that the theoretical and philosophical considerations which accompany the course shall follow a practical acquaintance with animals; rather than that special animal structures should serve chiefly as illustrative material for lectures on general biological theories, with a neglect of a thorough study of a series of animal forms. (c). The additional principles which should govern the planning of the introductory course, beyond those just stated, are: The selection of suitable teachers; The undesirability of attempting to establish a uniform preparatory course, or courses especially limited to applications to medicine; The acquirement of skill in the use of the microscope, and of correct scientific method of work in connection with the work of the course; The beginnings of embryology and cytology.

4. As to the last question, number 4, the report does not attempt to decide what proportion of the recommended preparation for anatomy can be obtained by a student in the first year's course. This must be indicated by the zoölogists. It seems evident to a student of present conditions, however, that most of the work desired in cytology and comparative, general histology; comparative anatomy of vertebrates; or systematic zoölogy will have to be elected by students looking forward to medicine, after they have taken the introductory course. It is to be hoped that the elements of vertebrate embryology will be included in that course. Some of this work may well be done in one of the excellent Summer Laboratories.

5. Finally, the importance of collateral reading in the masterpieces of biological literature is strongly emphasized.

H. McE. Knower, *Chairman*

On motion, the report as presented was adopted and the Committee was continued and instructed to confer with the Committee of the Zoologists on the basis of the report.

At the final session the Committee for Resolutions on the death of Professor Minot presented through its chairman, Prof. George S. Huntington, the following:

The American Association of Anatomists, assembled in the Thirty-First Session at St. Louis, record their profound sense of sorrow and their deep feeling of loss sustained by the death of Prof. Charles Sedg-

wick Minot of Harvard University. For many years Doctor Minot has stood for the best development of science and medical education both in this country and abroad. He has been particularly identified with the progress of the American Anatomical Association. In his official connections as President and Member of the Executive Committee his brilliant and constructive mind guided the affairs of the Society with marked success. Much of the progress of The Wistar Institute of Anatomy originated in his keen executive ability as Chairman of its Advisory Board. He was one of the founders of The American Journal of Anatomy and of its offspring, The Anatomical Record. His colleagues realize that these first American anatomical publications owed their success during their early and experimental years largely to his judgment and wisdom. Later he became most active in establishing and maintaining the eminently valuable relations now existing between the journals and the Publication Department of The Wistar Institute. These are mere outlines of a few of the more formal points of contact between Doctor Minot and the Anatomical Association. Important and far-reaching as these have been, and lasting as their impress will prove in the future development of the Society, they are fully equalled in value by the influence of his marked personality on the individual life and work of the members with whom he came in closer contact. Keen and yet considerate judgment, kindly help, both with advice and material, were freely extended by him. The Association, in mourning the loss of a stimulating leader, of a wise counsellor and of a personal force which has always directed forward the lone advance of national science, directs the following resolutions:

1. That the foregoing minute be published in the Proceedings of the Thirty-First Session and that the Secretary be requested to forward a copy to Doctor Minot's family.
2. That Prof. Frederick T. Lewis of Harvard University be requested to prepare, on behalf of the Association, a memorial of Doctor Minot's personal and academic life, with full consideration of his educational and scientific achievements, for publication in The Anatomical Record.

Before adjournment it was voted: That this Association extend a vote of thanks to Washington University, Professor Terry and his Staff, our hosts at this session, and of congratulations to them on the completion of their carefully planned and admirably equipped Institute of Anatomy.

CHARLES R. STOCKARD,
Secretary of the Thirty-First Session, of the American Association of Anatomists

ABSTRACTS OF PAPERS

1. The rhinencephalon of the dolphin [Delphinus delphis] William H. F. Addison, University of Pennsylvania.

In the adult dolphin, the olfactory bulbs and tracts are lacking, and that portion of the mesethmoid, which corresponds to the cribriform plate of the ethmoid of the ordinary mammal is imperforate. Thus the dolphin is entirely anasmotic, and it has been interesting to study the more centrally placed parts of the rhinencephalon, to see in them the extent of the regression which has accompanied the disappearance of the olfactory tracts and bulbs.

During the past summer, I had the opportunity of examining thin sections of the brain of an adult dolphin in the Frankfurt Neurological Institute, under the direction of Professor Edinger, to whom I am greatly indebted.

In addition to the lack of olfactory bulbs and tracts, the olfactory cortex of the basal surface of the frontal lobe is also wanting. At this region the corpus striatum of each side comes to the surface and protrudes as a convex oval area. This area, smooth and free from fissures, was named *lobe désert* by Broca. The parolfactory cortex is also much reduced, but at least some definite remains of it are seen. This is interesting in the light of Edinger's view, that the tuberculum olfactorium is not a part of the olfactory system, but is the end-station of tracts conveying impulses by way of the fifth nerve from specialized sensory structures in the snout region. To the sense, which this mechanism serves, he has given the name of 'oral sense.'

Of the several connections of the olfactory and parolfactory cortical cells with the hippocampus, only Zuckerkandl's bundle is definitely present. The fimbria is a slender band of fibers arising from the hippocampus. True fornix fibers are not seen, and in the usual region of the corpora mammillaria there are no well-developed rounded protuberances, and in the gray tissue of this region are seen no medullated nerve fibers. This would indicate that the tractus cortico-mammillaris is very small or perhaps lacking. There is the usual arrangement of a psalterium or crossing of fibers between the two hippocampi. Indeed, the psalterium is so well developed that the possibility is suggested that it may contain other fibers in addition to the commissural hippocampal fibers. The anterior commissure is much reduced, evidently the olfactory portion being entirely lacking.

Of the other possible connections of the olfactory and parolfactory cortical cells, both the taenia thalami and the taenia semicircularis are seen, as are their respective end-stations, the ganglion habenulae and the nucleus amygdalae. The hippocampi are very degenerate small

structures, and it is with difficulty that one sees the analogy with even the microsmatic type of hippocampus.

Thus, in the brain of the dolphin, accompanying the loss of the olfactory bulbs and tracts, there is found a recession of the frontal cortex, exposing the corpus striatum over a considerable area; loss of the olfactory portion of the anterior commissure; great diminution of the hippocampus, and reduction or loss of the uncrossed fibers from it to the corpora mammillaria (tractus cortico-mammillaris or true fornix); also, the usual connections between the olfactory cortex and the hippocampi are lacking except Zuckerkandl's bundle, and the corpora mammillaria are greatly reduced.

2. The artificial production of spina bifida by means of ultra-violet rays. W. M. Baldwin, Department of Anatomy, Cornell Medical College, New York City.

The purpose in presenting this paper is two-fold: first, by reason of the method employed the condition of spina bifida in tadpoles may be produced at will and the level of the bifurcation of the neural tube predetermined; second, the method gives considerable insight into the developmental potentials of the various portions of the ovum, and, in this instance, of the nature and location of the neural tube formative substances. This method consists in the illumination of small surface areas of the fertilized ovum of the frog by means of ultra-violet light of intensity sufficient to kill the area exposed in from 10 to 30 seconds.

It was found that in the undivided ovum the chemical organ-building substances, 'ferments,' or proanlagen, of the neural tube are neither located in any portion of the yolk hemisphere, nor along the equator. These proanlagen occupy a superficial position well up on the surface of the pigmented hemisphere of the egg and attain their definitive extent and position by a process of backward migration or differentiation, keeping pace in this migration with the corresponding shifting of the dorsal lip of the blastopore. These two processes occur synchronously, so that when the rate of the latter is interfered with (as from the presence of an area of dead yolk), the neural tube proanlagen differentiate into half anlagen and then half tubes at approximately their former rate of backward progression, but now along a line corresponding to the equator of the egg and not, as is usual, along the median plane of the egg. The yolk mass is finally completely drawn into the body of the embryo, and as a result the two neural-tube halves are approximated to a greater or less degree. Subsequent fusion of the tube-halves does not, however, occur. Each half-tube by a later shifting of its cell becomes a whole tube.

The experiments add one more fact towards the establishment of the conception of the egg as a composite structure containing, from the first, the organ-building substances of the various body systems, restricted to more or less definitely localized areas in the egg substance (mosaic theory of Roux). Furthermore, the conclusion seems justifiable, tentatively, at least, that the various chemicals such as salts of sodium and of lithium, which have been used by Morgan, Hertwig.

Herbst, Jenkinson and others in the production of this malformation, have, at least, produced their effect by acting upon portions of the yolk hemisphere and not necessarily upon the proanlagen restricted to the pigmented hemisphere.

C. R. Bardeen, see abstract 57, page 137.

3. Some effects of mammalian thyroid and thymus-glands upon the development of Amphibian larvae. G. W. Bartelmez, Department of Anatomy, The University of Chicago.

The following data are taken from some attempts made with a view to analyzing the reactions reported in the feeding experiments of Gudernatsch and others. They are based on experiments upon larvae of Amblystoma tigrinum and Rana catesbiana treated with sheep's thyroid and thymus in three different ways, appropriately controlled. (1) Using the glands as food. (2) Feeding normally but adding saline extracts of the glands to the water of the culture dishes. (3) Injecting strong extracts into the coelom or dorsal musculature.

Experiments with thyroid gland. Amblystoma: Effects of feeding (spring, 1913). Amblystoma is a favorable form in that the larvae can be hatched and reared in the laboratory, that they are fairly hardy, more especially that they are carnivorous and the effects of feeding can be studied separately from those resulting from suspensions of the glands in the culture water. When exclusive thyroid feeding was begun soon after hatching (12 to 15 mm. larvae) there was a high mortality but the survivors showed only the effects of a meagre diet such as was obtained by feeding egg albumen or by starving. They grew little or not at all and the limbs did not differentiate as rapidly as in the controls. No clear cut results were obtained unless the larvae were at least 30 mm. long, had three or four fingers and leg buds. In these cases a few individuals survived to undergo partial metamorphosis. Still older larvae, if they were well nourished, after two or three feedings of thyroid underwent metamorphosis in the course of eleven to sixteen days. Fairly normally constituted adults were thus obtained from 35 to 80 mm., long whereas the normal in this vicinity at the time of metamorphosis is from 130 to 150 mm. long. My conclusions from these observations are that the thyroid feeding does not stimulate differentiation in Amblystoma since the differentiation of the limbs is not accelerated but it does bring about certain changes in the gut which in turn induce other changes characteristic of metamorphosis.

Amblystoma: Effects of thyroid extracts (spring and summer, 1914). In these experiments the evil effects of an unnatural food were eliminated by giving the larvae first entomostraca and then anuran tadpoles as food and between the feedings adding the extract to the water in which they were living. Starting with larvae 15 to 17 mm. long, after six treatments in the course of six weeks no differences were noted as compared with the controls. During this time they grew and differentiated normally, reaching a length of from 20 to 40 mm. Continuing the treatment for two and a half months, small adults were obtained. Begin-

ning with older larvae the process was somewhat more rapid. The chief differences between these and the feeding experiments lie in the lowered mortality and the more complete metamorphoses in the former set. Both agree in that the thyroid has no specific effect until the larva has reached a definite minimal stage in development and in that the metamorphosis is not wholly normal.

Amblystoma: Effects of hypodermic injections (summer, 1914). The results of injecting small doses of strong extracts of thyroid were somewhat variable, largely no doubt because in different cases different proportions of the injection oozed from the wound. Animals under 50 mm. in length gave no signs of metamorphosis before death. In older ones two doses were sometimes necessary to start the process and then it began within four to seven days and was completed in fourteen to thirty days: a period somewhat longer than normal.

Rana catesbiana: Experiments with thyroid gland. The results with bull frog tadpoles were complicated by various factors and only a few experiments can be summarized here. The reaction varied according to the stage of development of the larva, its age (1st, 2nd or 3rd season), the length of previous confinement in the laboratory and the season of the year in which the treatment was begun. The individual resistance was also variable and this was a factor as the supply of material was limited and each tadpole was measured and observed until its death.

Effects of feeding. Larvae fed only twice with thyroid and the rest of the time with lymph node reacted like those fed exclusively on thyroid and as the death rate was lower, the former treatment was used in most cases. In larvae of all ages five or six days after the first feeding the body became more slim than in the protein fed controls. This was due to the marked reduction in the length and in the position of the spiral gut. After this differences were noted which depended upon the stage of development reached by the animal at the time thyroid feeding was begun. If the legs had developed so far that the toes were differentiated at time of first feeding, the legs grew as they do shortly before metamorphosis, but true metamorphosis did not set in until after six or seven weeks. Some of this group, however, developed a marked resistance to the thyroid. To cite a single case: An individual which had been accustomed to a lymph node diet and then to thyroid by six feedings between January 19 and April 7, was fed weekly with thyroid until May 21 (seven times) then on alternate days twelve times, died half way through metamorphosis on June 16. In this and the cases mentioned above the gut reduced at a faster rate than it does normally. This fact is accentuated by the following class of cases. When thyroid feeding was begun before the larva had gone beyond the stage of leg buds a peculiar kind of metamorphisis was brought about. The gut shortened and assumed practically the adult condition, the head showed some signs of metamorphosis but there was no reduction of gills, little reduction of the tail and no more development of the leg buds than in the control.

The results of treatment with thyroid extract and the injection of thyroid suspensions in general confirmed these results with feeding.

Upon the assumption that lymph node tissue is similar as a food to thymus, but that it has no internal secretion, some series of experiments were made with these two tissues using them both as foods and as extracts in the culture dishes. In Amblystoma they gave practically identical results in all feeding experiments—and there was no proof of a retardation of development. Larvae treated with thymus extract metamorphosed normally but sooner and when the animals were smaller than the controls. Furthermore, the hypodermic injection of thymus extract did not retard or inhibit metamorphosis in the larger larvae. In Rana the feeding of both thymus and lymph node produced more rapid development than was observed in the plant fed control.

4. The development of the sympathetic nervous system in Elasmobranchs.

Geo. A. Bates, Tufts College Medical School, Boston, Mass.

The first appearance of the sympathetic in Squalus is in the form of a series of ganglia lateral to the aorta in 15 mm. embryos. At the time of its formation the dorsal and ventral roots of the somatic spinal nerves, in their ventral growth, have reached this level. The ganglion of the sympathetic is formed in immediate connection with the dorsal root, from cells that arise from this source. It appears as a cluster of cells attached to the dorsal root and embedded in a protoplasmic mass quite distinct from the surrounding mesenchymal cells.

In embryos of from 7 to 8 mm. cells of the ventro-lateral wall of the neural tube have begun to migrate into the space between the tube and myotome. At the same time mesenchymatous cells from the disintegrating sclerotome migrate dorsally into the same region. The medullary cells are easily distinguishable from the sclerotomic cells in sections prepared by the vom Rath method. These cells arrange themselves along the margin of the myotome and are distinctly marked off from the surrounding cells.

There are no medullary cells among the cells of the mesenchyma. In the latter, however, two varieties of cells are present; one reacting to the basic stain quite intensely, the other less so. These facts are demonstrated in sublimate fixed, hematoxylin stained sections, and particularly in sections stained with iron hematoxylin.

Such cells are present throughout the mesenchyma and at points where neither the ventral root cells nor cells from the dorsal crest have begun their migration.

The claim that the deeply staining cells in the mesenchyma are medullary cells which subsequently will become incorporated into the sympathetic ganglion, seems unwarranted for the following reasons:

At this stage, 7 to 8 mm. there is no sign of the formation of the sympathetic ganglion. At the level of the aorta and lateral to it a mass of sclerotomic cells may be seen and it is here that the two sorts of cells, above mentioned, are found.

The difference in staining property seems to be the result of the presence or absence of chromatin due, probably, to the state of activity of the cell. Such conditions have frequently been observed in mesodermic cells in the formation of the liver, and also by various observers in other regions. In other words, the difference in staining properties of cells in the region of the dorsal aorta affords no foundation for the inference that the more deeply staining cells are destined to become sympathetic cells. On the contrary, they are mosodermal in their origin and are not genetically related to the sympathetic.

As above stated, the sympathetic ganglion is developed directly from the dorsal root of the somatic spinal nerve at a stage of about 35 to 17 mm. At the time of development there are relatively few cells present in the motor root, and the question of contribution to the ganglion of cells from that source, while not improbable, is doubtful.

5. The growth of the head and face in American (white), German-American, and Filipino children (lantern and photos). ROBERT BENNETT BEAN, The Tulane University of Louisiana.

Materials:

146 Filipino girls 579 Filipino boys	Manila, Philippine Islands
309 German girls 324 German boys 412 American girls 415 American boys	Ann Arbor, Michigan
2185, total	

The growth of the head diameters (length, breadth and height). Between the ages of 6 and 16 the head grows in length least, 0.9 cm., in the American girls, and most, 1.6 cm., in the Filipino boys; in breadth least, 0.5 cm., in the American girls, and most, 1.1 cm., in the German boys; and in height least, 0.5 cm. in the German and American girls, and most 1.1 cm. in the Filipino boys. The heads of the Filipinos grow more rapidly in length between 6 and 11 years of age than between 11 and 16 years of age, whereas the heads of the Germans and Americans grow more rapidly in the latter than in the former period. What is true of the Germans and Americans in relation to the Filipinos is also true of the boys in relation to the girls.

The head size as represented by the module (length plus breadth plus height) increases least, 19 points, in the American girls and most 35 points in the Filipino boys.

At 6 years of age the heads of the Americans of both sexes are the largest, the heads of the Filipinos are the smallest, and the heads of the Germans are nearly as large as those of the Americans. At 16 years of age the heads of the Filipinos are the smallest, and the heads of the Americans are nearly as large as those of the Germans.

The cephalic index decreases with age for the length-breadth index least, 0.0, for the Filipino girls, and most, 3.3, for the Filipino boys;

and for the length-height index least, 0.4, for the Filipino boys, and most, 2.7, for the German girls.

Growth of head circumferences (frontal, forehead, parietal and occipital.) The forehead and occipital regions are large in the boys and in the Americans, the frontal and parietal regions are large in the girls and in the Germans and Filipinos. The forehead and frontal regions together are large in the girls and in the older children, and the occipital and parietal regions together are large in the boys and in the younger children.

From 6 to 16 years of age, the forehead, frontal, and parietal regions grow most in the Filipinos, less in the Germans, and least in the Americans, but the reverse is true of the occipital region. The forehead, frontal and occipital regions grow more in the boys than in the girls, and this is especially true of the occipital region, whereas the parietal region grows more in the girls than in the boys.

It is notable that, in relation to each of the other regions, the forehead increases in size and the parietal region decreases with age.

The large size and greater growth of the parietal region are characteristic of the girls and of the young children, and the large size and greater growth of the occipital region are characteristic of the boys and of the older children. The Filipinos resemble the girls in this respect, and the Americans resemble the boys, whereas the Germans are more or less intermediate.

The Hypo-types are like the Filipinos, the Hyper-types are like the Americans and the Meso-types are like the Germans.

The growth of the face (length, breadth and facial angle): The growth of the face as a whole may be considered by taking the product of the length and breadth. From this standpoint the growth from 6 to 16 years is least in the Filipino girls, greatest in the American boys, with the others in between, the boys greater than the girls. The face increases about 33 per cent in the girls and about 50 per cent in the boys during the ten year period.

The face length increases with age about 2 cm. in 10 years. The girl's face grows more from 6 to 11 years and the boy's from 11 to 16 years. The face of the Filipino is shorter than that of the German and American, about 1 cm. at 16 years and about 0.3 cm. at 6 years. The face of the Filipino grows less in length than that of the German and American from 6 to 16 years.

The face breadth increases with age from 11.3 cm. at 6 years to 13.1 at 16 years. The face breadth of the girls grows more rapidly from 6 to 11 and that of the boys from 11 to 16. The face of the Filipino is as broad as that of the German and broader than that of the American, and the growth of face is about the same in breadth for the three peoples.

The face index increases with age, the face becomes longer relative to its width, and this increase is greatest in the Americans, less in the Germans, and least in the Filipinos. The increase in the Germans is greatest from 6 to 11 years and in the Americans it is greatest from 11 to 16 years.

The facial angle represents the projection of the maxilla, and with

increase of age this is greater in the American boys and less in the Filipino boys than is apparent in the German and American girls and the German boys. The Filipino girls have no records made of the facial angle.

Cephalo-facial index (originated by the author). This represents the size of the face in terms of the head, the latter always 100. The face grows relatively more than the head from 6 to 16 years, relatively more from 6 to 11 in the Germans and Americans and relatively more from 11 to 16 in the Filipinos. At 6 years the Filipinos have relatively the largest faces, and the Americans relatively the smallest, with the Germans in between, at 11 this is reversed, and at 16 all are about the same.

The cephalic index decreases with age, and it decreases from the Filipinos through the Germans to the Americans. The face index increases with age, and it increases from the Filipinos through the Germans to the Americans. If the process of development recapitulates the progress of evolution then the Americans represent in evolution what the adult represents in development, and the Germans and Filipinos are less mature stages. The Filipinos represent what I have called Hypo-phylo-morphs, the Germans Meso-phylo-morphs (?) and the Americans Hyper-phylo-morphs. In each group may be found adult individuals with varying degrees of development in head and face form, and these I would classify as Hypo-onto-morph, Meso-onto-morph, and Hyper-onto-morph, depending upon the extent of development. Crossing of races has introduced the phylo- types into nearly all peoples, therefore the six forms may be distinguished among almost all mixed races. Among the white peoples the Hypo- types are rare, but among the Filipinos the Hyper- types are abundant. More white peoples have mixed with the Filipinos than Filipinos with the white peoples.

6. Some ears and types of men. (*Lantern and photos*). ROBERT BENNETT BEAN, The Tulane University of Louisiana, New Orleans, La.
Materials:

1325	American whites
2039	American negroes
73	American Indians
171	Alaskan Eskimos
94	Manila Filipinos
3702	Total

The present study is a continuation of those made previously on the external ear and physical form of man, and it is more detailed and specific than former studies. It corroborates them in general and in particular, and adds racial distinctions to type differences.

The most important result is the segregation of the Hyper-, Meso-, and Hypo- types from each group, both by the ear form and by other anatomical characteristics. The other result of importance is the differentiation of the races by their ear form. Incidentally skin lines

were discovered on all ears, lines that represent the folded over skin tip of the ear, the skin tip which should overlie the cartilaginous tip (Darwin's tubercle) but does not always do so.

The segregation of the types, Hyper-, Meso-, and Hypo-, is accomplished by determining for each ear whether the helix is prominent or not, whether the anthelix is depressed or not, whether the lower helix and lobule turn towards the head or away from it, and whether the tragus and antitragus are everted or depressed. After having determined to which type the ear belongs, then the cephalic index, nasal index and facial index of the individual are calculated. The results are found below.

Type differences. Hyper-: In this type of ear the helix is depressed toward the head, the anthelix is prominent—projects beyond the helix—the lower helix and lobule turn toward the head, and the tragus and antitragus are everted and prominent—project beyond their surroundings. The nasal index, facial index and cephalic index indicate that the type of individual associated with this type of ear has a long, narrow nose, a long, narrow face, and a long narrow head as a rule.

Hypo-: In this type of ear the helix is prominent, the anthelix depressed, the lower helix and lobule turn out from the head in the form of a shelf, and the tragus and antitragus are depressed below their surroundings. The nasal index, facial index, and cephalic index indicate that the type of individual associated with this type of ear has as a rule a short, broad nose, a short broad face, and a short broad head.

Meso-: In this type of ear the helix and anthelix are both prominent, thus forming a double roll near the dorsal margin of the ear, the lower helix and lobule turn out from the head in the form of a shelf, but not to the same extent as in the Hypo- ear, and the shelf, instead of being horizontal, has a gentle slope forward or may be precipitous, and finally the tragus and antitragus have an intermediate position, are neither prominent nor depressed. The nasal index, facial index, and cephalic index indicate that the type of individual associated with this type of ear has a nose, face, and head of intermediate form between the Hyper- and the Hypo-, although the face is larger than either of the two.

Each of the three types may be subdivided into onto and phylo forms, the phylo, the primordial form, and the onto, the derived form. The Hyper-onto-morph, the Meso-onto-morph, and very rarely the Hypo-onto-morph are European, or white, types; whereas the Hypo-phylo-morph, the Meso-phylo-morph, and rarely the Hyper-phylo-morph are types of the negroes, Indians, Eskimos, Filipinos, and other primitive peoples.

At birth the white child is a Hypo-phylo-morph, and as the child develops it passes consecutively through the stages of the Hypo-onto-morph, Meso-phylo-morph, Meso-onto-morph, Hyper-phylo-morph and Hyper-onto-morph, unless development stops at or between one or the other of the types.

There is little doubt that the Hyper-ontc-morph is the end product of a hyperactive thyroid gland, the result of rapid differentiation, with slight growth, resulting in a small, active, nervous individual. The Hypo-phylo-morph is probably the end product of great thymus activity, resulting in a more or less complete retention of the infantile condition, whereas the Meso-phylo-morph has great activity of the gonads. The other types are variants of the three mentioned, composites, mixtures, blends or mosaics.

Race differences. The race differences are of two kinds, measured and descriptive. Only the racial differences of the ear will be considered here.

Measured differences: These are divided into differences in the living and differences in the dead. The ears of only three groups of dead people were measured, American negroes, American whites and Filipinos. By measurements of the total ear length, total ear breadth,

TABLE 1

GROUP	EAR LENGTH		EAR INDEX	
	Male	Female	Male	Female
Dead				
American white	64.18		58.0	
American negro	58.58	58.32	64.0	60.8
Manila Filipino	58.80	57.43	56.8	57.5
Living				
New Orleans student	63.9		57.4	
American "old" white[1]	66.9	61.3	55.9	56.0
American Indian	72.3		52.6	
Alaskan Eskimo	73.8	67.1	54.4	53.0
American negro	60.8	58.0	60.9	60.2

[1] 'Old' whites are those who have been in this country for 3 generations or more.

Foetuses, new-born and young infants, male and female:

Ear index white	68.8
Ear index negro	64.5

ear base, true ear length (Schwalbe), concha length and concha breadth, it is found that the negro ear is short and broad, the white ear is long and narrow, and the Filipino ear is relatively longer and narrower than the white ear. The ear length and the index of the ear of both the living and the dead are shown in table 1.

No other measurements are given because racial differences are more pronounced in the ear length and the ear index. The Indian and Eskimo have long ears, the negro and Filipino have short ears and the ears of the white people are intermediate. This accounts in part for the fact that the ear index of the negro is high, that of the Indian and Eskimo is low, and that of the white is intermediate, but it

does not account for the low index of the Filipino. The reason for this is that ear of the Filipino is short and also narrow, it is a small ear. The negro ear is not only short, but it is also broad.

The ear increases in size with age, to 70 years or later, but the increase in length is greater than that in breadth, therefore the index decreases with age.

Descriptive Differences: The true negro ear is small, almost flat, close to the head, and the helix is broad as if much folded over. The upper part of the helix is almost horizontal and passes directly backward from the upper end of the ear base to join the vertical dorsal portion of the helix at a right or acute angle in a rounded point at the upper outer extremity of the ear. The superior and dorsal borders of the helix are separated by a depression above Darwin's tubercle, where the helix is thin or absent. The dorsal border passes downward and turns forward at an obtuse angle to form the inferior border of the ear which enters the cheek almost at right angles, with no lobule or a very small one which is nearly flat. The Satyr tubercle is well marked and Darwin's tubercle is small or absent. The skin lines formed by the overfolding of the helix are less distinct on the negro ear than on the white, and they usually converge on the negro ear over Darwin's tubercle. The true negro ear is not seen in great numbers among American negroes. It occurred 245 times among 1478 New Orleans negroes (16.6 per cent), men, women and children, chiefly of the laboring classes.

There is another form of ear that is found frequently among the negroes, but it is also found not rarely among other peoples, even among the whites, and I have called this the involuted ear, because it seems to represent an advanced stage in retrograde development and evolution. It has a gnarled appearance, as if the ear had been burned around the border and had contracted irregularly in healing, leaving a thick, irregular helix. This ear type was at first thought to be due to accidental causes, but the presence of the skin lines of the ear tip in regular order proved the ear to be a true type. It was found 601 times in 1478 New Orleans negroes (40.7 per cent) and 52 times among 857 New Orleans whites (6.1 per cent).

The details of the ears of the negro and white are different as follows: The negro ears are glabrous, the white ears are hirsute; the Satyr tubercle is large in the negro ear, small in the white; Darwin's tubercle is more difficult to find in the negro ear than in the white; the skin lines converge about Darwin's tubercle in the negro ear, and between Darwin's tubercle and the Satyr tubercle in the white; the helix is broad in the negro ear, narrow in the white; the anthelix is more prominent in the white ear than in the negro; the posterior auricular sulcus is deeper in the negro ear than in the white, and in the negro ear the sulcus dips into the concha, whereas in the white it turns out over the helix or lobule.

I wish to thank Dr. Hrdlička for some of the records of the American whites and negroes and for the records of the Alaskan Eskimo and American Indians.

7. Absence of the vena cava inferior in a 12 mm. pig embryo, associated with the drainage of the portal system into the cardinal system. ALEXANDER S. BEGG, Harvard Medical School.

In a 12 mm. pig embryo which appeared normal before being sectioned, I found very radical anomalies of the abdominal veins. The vena cava inferior, which arises through anastomosis of the right subcardinal vein with the sinusoids of the liver in pig embryos of about 6 mm. and which is very large in 12 mm. embryos, had failed to develox. Thus this embryo presented a young stage of the interesting and well-known anomaly of the adult, described as 'absence of the vena cava inferior.'

Moreover, the portal system did not empty into the liver by the usual large venous trunk, but only through capillary connections, very difficult to follow. On the other hand, the connection between the portal system and the cardinal system, which is ordinarily insignificant at this stage, is an important, if not the chief, outlet of the portal vein. Thus this embryo shows in an early stage the rather rare anomaly of the adult in which a large vein passes from the splenic vein to the left renal vein.

In order to show accurately these features, models have been made of the vessels in the abnormal pig and also in a normal specimen for comparison. Except for the decrease in the size of the portal vein as it enters the liver, the abnormalities are a persistence of earlier normal conditions.

8. Notes on the endocranial casts of Okapia, Giraffa and Samotherium DAVIDSON BLACK, Anatomical Laboratory, Western Reserve University, Medical School.

The superficial convolutional pattern of the convex surface of the cerebrum in the lower gyrencephalous mammalia, more especially in the Ungulates and Carnivores, is quite accurately reproduced by the corresponding irregularities of the internal surface of the skull. The major portion of the lateral surface of the neopallium is exposed in these forms—more especially in the Ungulata. In other words, no very extensive operculae are present to obscure the fundamental fissural pattern, which may thus be accurately studied in the endocranial cast.

One endocranial cast of an adult Okapia and one of an adult Giraffa camelopardalis were obtained in Manchester through the courtesy of Prof. G. Elliot Smith. The second specimen of an adult Okapia, together with that of Samotherium, was obtained through the courtesy of Dr. Smith Woodward from the Museum of Natural History, at South Kensington, London. I am also indebted to Prof. Arthur Keith for the opportunity of studying the giraffe brains in the collection of the Royal College of Surgeons, and to Dr. C. U. Ariens Kappers for the privilege of studying the Ungulate and other material in the collection of the Central Dutch Institute for Brain Research in Amsterdam.

These notes, together with the illustrations here shown as lantern slides, will form the basis of a more extensive paper in the near future.

Giraffa: The convolutional pattern in the giraffe is well known from the study of actual specimens of the brain, and may be readily outlined upon the surface of the cast. Dorsally the lateral sulci and suprasylvian arc are well marked. The coronal sulcus shows a caudal connection with the ansate sulcus, which is a common ungulate condition. This 'ansata' is in no way to be compared with the somewhat similarly placed cruciate sulcus peculiar to the carnivores. The olfactory bulbs are only seen in this view as small swellings projecting very slightly from beneath the frontal pole.

The whole course of the suprasylvian sulcus is shown in the lateral view of the cast. In addition to the post. horizontal limb common in Cervidae and other forms, a posterior descending ramus of the suprasylvian sulcus is to be noted. This sulcus is probably not homologous with the postsylvian sulcus of the carnivores. The posterior rhinal fissure is well marked and above it can be seen the bulging of the 'post. sylvian operculum,' the edges of which are indented by a series of small sulci.

The Ungulate sylvian, or pseudosylvian, fissure appears as a short ascending ramus from the diverging ectosylvian sulci. Between the summit of this pseudosylvian fossa and the suprasylvian sulcus there is seen a well marked arcuate fissure. This 'arcuate' constellation is in no way homologous with the ectosylvian group so evident in Canis, Felis, etc., but appears to be a characteristic feature of the giraffidae, and when present together with the posterior descending ramus of the suprasylvian, is a distinct diagnostic feature.

The anterior rhinal fissure, together with the orbital and paraorbital sulci present no peculiarities. There is a typical triradiate diagonal sulcus. Posteriorly, the area behind the descending ramus of the suprasylvian sulcus is marked by several irregular sulci, as is the case in the corresponding area behind the oblique sulcus in Cervus.

The cast is one of a typically macrosmatic mammal of the Ungulate type. The olfactory bulbs are very large and sessile and, together with the tractus olfactorius and tuberculum, are best seen in the ventral view. Here, as also in the lateral view, the enormous size of the combined ophthalmic and maxillary divisions and also the large mandibular divisions of the trigeminal nerve is at once evident, the optic nerves being small in comparison. The large size of the N. V. is directly related to a highly developed so-called 'oral sense.'

Okapia: This animal is a hornless member of the family Giraffidae, and was first discovered in the Belgian Congo in 1899 by Sir Harry Johnston. No material other than the skin and skeleton has as yet been available for scientific study.

The arrangement of the sulci appearing on the dorso-lateral surface of this cast gives evidence of a close relationship between the Okapia and the Giraffe. The 'arcuate' constellation, the descending ramus of the suprasylvian and the position occupied by the lateral group of sulci are essentially similar to those obtaining in the giraffe.

There are, however, numerous specific differences. The descending ramus of the suprasylvian fissure cuts the post. rhinal fissure in Okapia.

In Cervus dama and many other forms, an 'oblique' sulcus occupies the area in which this descending ramus is found in the Giraffidae. This oblique sulcus in Cervidae in many cases cuts the rhinal fissure, and it may be that this sulcus is the homologue of the descending ramus of the suprasylvian. Thus the cutting of the post. rhinal fissure by the latter sulcus may be a premature feature common to Okapia and Cervus, but absent in the more specialized giraffe.

The pseudosylvian fissure is very short and the relation of the anterior ectosylvian sulcus to the anterior rhinal fissure is obscure. The irregular sulci in the area behind the ram. desc. of the suprasylvian sulcus present essentially similar relations to those in the giraffe.

The relations of the coronal, ansate and suprasylvian sulci together with the diagonal are quite different to the conditions obtaining in the giraffe. The suprasylvian is joined to the corono-ansate sulci, as is the case in the Cervidae and often in the other Ungulates. The absence of this condition in the giraffe may thus be considered as another evidence of specialization in this form.

A constellation apparently representing the 'diagonal' sulcus of Elliot Smith appears continuous with the suprasylvian (on the right side in the specimen illustrated). On the left side of the same specimen the diagonal sulcus occupies its usual position in front of and below the coronal sulcus.

The olfactory bulbs are large and pedunculated, and project a considerable distance beyond the frontal pole, so that the olfactory stalks are visible in the dorsal view. The olfactory tracts and tubercule, together with the pyriform lobe, are well seen in the ventral view. The tuberculum is especially prominent and occupies a special little fossa on the skull floor. The total amount of neopallium as compared with the rhinencephalon appears less than in the case of the giraffe.

Samotherium: This animal was an Okapi-like form found in the upper Miocene deposits in the island of Samos. The cast shows evidence of considerable compression during fossilization, resulting in marked asymmetry. Notwithstanding this unfavorable condition, it is possible to trace the course of the principal sulci with but little difficulty.

The entolateral sulcus occupies its usual position, but the lateral sulcus takes a very unusual course and becomes joined to the coronal as well as with the suprasylvian, through the intermediation of the ansate sulcus. This junction of lateral and coronal is found nowhere else in the Ungulata except in the primitive hippopotamus. In other orders, however, as for example in the Carnivora, this junction between coronal and lateral sulci is a common feature. Elliot Smith has shown this feature to be a very primitive character and present in such Eocene Carnivores as Stenoplesicites and Gynohyaenodon as well as in ancestral Ungulates.

The continuity of the suprasylvian and coronal by way of the ansate sulcus is, as has been already noted, a common feature in Ungulates, but is practically never present in Carnivores.

The coronal sulcus is placed far forward, and is comparatively small as in Hyrax, while the ansate sulcus is well developed. In front of the coronal sulcus a triradiate diagonal fissure is evident in its usual position.

The orbital sulcus emerges from the anterior rhinal fossa and is placed far forward, as in many of the Cervidae and Suidae.

A sulcus recalling the Carnivore 'cruciatus,' but not to be homologized, appears emerging from the sagittal furrow and probably represents the upturned termination of the splenial.

The ramus descendens of the suprasylvian sulcus cuts the posterior rhinal fissure as in Okapia. The pseudosylvian sulcus on the left side is represented by a series of small vertical notches, the whole being related to a typical 'arcuate' sulcus, such as obtains in Okapia and Giraffa. The pseudosylvian sulcus on the right side more nearly approaches the condition obtaining in Giraffa.

Summary: From a study of the limited amount of material at my disposal it appears that of the three Artiodactyl forms of the family Giraffidae under discussion, the brain of Samotherium shows undoubtedly the most *primitive* arrangement of sulci, presenting as it does certain features common both to the Carnivora and the Ungulata. In other words this form is evidently most closely related to that hypothetical 'co-mammal' from which both the Carnivora and Ungulata were specialized.

In addition to this primitive feature, Samotherium presents certain *generalized* characters, common and peculiar to the Ungulata such for example as (a) the relation of the descending ramus of the suprasylvian sulcus to the post. rhinal fissure (provided the former sulcus be the homologue of the 'oblique' sulcus of Elliot Smith) and (b) the arrangement of the coronal, ansate, suprasylvian complex anteriorly. In these generalized characters Okapia resembles Samotherium and the two differ from Giraffa.

All these forms show a certain fundamental similarity in fissural pattern. These *specialized* characters are seen in the arrangement in the 'intrasylvian arcuate' complex which, taken in conjunction with the descending ramus of the suprasylvian sulcus is apparently peculiar to Giraffidae.

And finally, Giraffa differs from both Samotherium and Okapia in the possession of certain specialized features, such, for example, as the complete separation of the corono-ansate group from the suprasylvian sulcus.

9. Explanation of variations of the renal artery. J. L. Bremer, Harvard Medical School, Boston.

Vessels mentioned in the text-books of anatomy as either anomalous roots or anomalous branches of the renal artery may be placed in three groups: (1) those to the mesonephros and its adjacent organs, ventrolateral branches of the aorta; (2) those to the intestinal tract and its derivatives, ventral branches; and (3) those to the body wall and dia-

phragm, dorso-lateral branches. Group 1 includes the spermatic and adrenal arteries, and branches of the iliacs and middle sacral; group 2, the coeliac axis and its branches to liver, pancreas, and colon, and the superior and inferior mesenteric arteries; and group 3, the lumbar arteries and the inferior phrenic. If horizontal anastomoses between members of the different groups can be found, and if in addition longitudinal or vertical anastomoses between members of the same group exist, any of the variations are explicable.

At the outset it was found that, whereas in man the renal artery is normally a branch of a mesonephric artery, in pig and sheep the new vessel is derived from body wall (or lateral body) vessels.

The origin of the renal artery in man from the iliacs or the middle sacral is due to the original pelvic position of the kidney, in the immediate vicinity of the vessels mentioned. Branches from them, similar to mesonephric arteries, may run to the kidney, and may continue in activity as the kidney migrates, instead of being lost or becoming ureteric arteries, as is more usual. Spermatic and adrenal branches of the renal artery are not uncommon, and are due to the fact that all are derivatives of the mesonephric arteries. Vertical anastomoses of mesonephric arteries are common, and since now one root, now another, is kept, such anastomoses account readily for the frequent asymmetrical origin of the renals from the aorta. Vertical anastomoses between dorso-lateral aortic branches are also common in the abdominal, as well as in the cervical, region.

Horizontal connections, rarer than the vertical, are found oftenest between the ventral and the ventro-lateral group, and account for the origin of the renal artery from the coeliac axis or the mesenteric arteries, and for the branches of the renal to liver, pancreas, and colon. A double anastomosis between one of the early ventral arteries and a mesonephric artery on each side, with the subsequent loss of the ventral artery and of any two of the three roots to such an anastomosis, would result in the origin of the renals from a common stem.

Horizontal anastomoses between vertebral, or dorsal, and lateral body arteries can hardly be considered anomalies, as in most animals the two sets of vessels soon come from a common stem, as in adult man. Horizontal anastomoses between lateral body and mesonephric arteries are very rare, but serve to explain the origin of the renal artery in man from a lumbar artery, or the presence of a phrenic branch of the renal.

The various anastomoses found may be so close to the aorta, when it is only an endothelial tube, that they become incorporated in its wall by the development of the muscular layer.

It will thus be seen that all the variations of origin or anomalous branches of the renal artery can be explained by the presence, in different embryos, of pieces of a periaortic anastomoses, joining the various aortic branches, vertically and horizontally. If these pieces were all developed in one individual, it would be capable of transferring the blood stream from a dorsal vessel to a ventral one, and vice versa. In

this connection it is interesting to note that the mesonephric arteries of adult selachians are said to spring normally from the segmental body wall vessels, and that in Bdellostoma the mesenteric arteries arise from the dorsal wall of the aorta.

10. Comparative size of nucleus and cytoplasm in old and regenerating tissues. E. L. BREZEE, Cornell University Medical School, New York City. (Introduced by C. R. Stockard.)

The species used for this experiment were Fundulus heteroclitus, tadpoles of Rana sylvatica, and two species of salamander, Diemyctylus viridens adults and Amblystoma punctatum larvae.

Old and regenerating tissue from both adult and larval animals were studied. The tails and arms were cut off about one-third or one-half way from the body. These parts after a few days had regenerated and the organisms were then preservd in Bouin's fluid, sectioned and stained with hämatein and eosin.

Sections were cut in such a plane as would pass through both old and regenerating tissue. Two sections of each specimen were studied and camera lucida drawings made of the nuclear outlines of portions of epithelium and mesenchyme from the old and new tissue of each section. The cell outlines could not be traced as they did not show distinctly enough. In the epithelium the cells were so closely packed that all the substance which was not nucleus could be considered as cytoplasm. In the mesenchyme, however, the cells were so scattered and the intercellular spaces so numerous that the amount of cytoplasm could not be determined in this way; hence no relation between the amount of nuclear material and of cytoplasm could be calculated, but merely the size of nuclei in old and new tissue compared.

The purpose of the experiment was three-fold: (a) to determine whether the nuclei of the old or of the new tissue were larger; (b) to determine in both old and new tissue whether the amounts of nuclear or of cytoplasmic material were greater; and (c) to compare the relative amounts of nuclear and cytoplasmic material in old and new tissue. In each case the areas traced from the old and new tissue of the same section were compared, and the results noted.

(a) By comparing the tracings from each portion of new tissue with the corresponding portion of old it was possible to determine without actual measurement in which the nuclei were larger. The results are shown in table 1.

In the mesenchyme there is a slight advantage in nuclear size shown in the old tissue; this is true also of the epithelium of the larvae but in the adult animals the size of the nuclei of the new epithelium is more often greater.

(b) The actual areas of nuclear and cytoplasmic material were found by the following method: a rectangular portion was outlined and its area found; the mean diameter of each nucleus within this portion was determined and its area found by use of the formula πr^2, letting $\pi =$

3 $\frac{1}{7}$. The sum of these areas gave the total area of the nuclei and by subtracting this from the area of the rectangle, the area of the cytoplasm was found. Then, using the ratio

Area of nucleus : Area of cytoplasm : : 1 : X

the number of parts of cytoplasm to one part of nuclear material was found for each specimen. In the majority of all cases the area of cytoplasm was greater than of nuclear material; i. e., in the ratios X was found to be greater than 1. Table 2 shows the number of cases which were exceptions:

TABLE 1

SPECIES	NUCLEI LARGER IN OLD TISSUE AS COMPARED WITH NEW	EQUAL	NUCLEI LARGER IN NEW TISSUE AS COMPARED WITH OLD
Mesenchyme			
Adult			
Fundulus heteroclitus tail	2 cases	4 cases	4 cases
Diemyctylus tail	31 cases	18 cases	15 cases
Diemyctylus arm	5 cases	3 cases	12 cases
Total	38 cases	25 cases	31 cases
Average	40 per cent	27 per cent	33 per cent
Larval			
Rana sylvatica tail	6 cases	2 cases	2 cases
Amblystoma punctatum tail	10 cases	13 cases	7 cases
Total	16 cases	15 cases	9 cases
Average	40 per cent	37½ per cent	22½ per cent
Adult and larval			
Total	54 cases	40 cases	40 cases
Average	40 per cent	30 per cent	30 per cent
Epithelium			
Adult			
Fundulus heteroclitus tail	7 cases	No cases	3 cases
Diemyctylus tail	21 cases	12 cases	31 cases
Diemyctylus arm	2 cases	2 cases	16 cases
Total	30 cases	14 cases	50 cases
Average	32 per cent	15 per cent	53 per cent
Larval			
Rana sylvatica tail	4 cases	2 cases	4 cases
Amblystoma punctatum tail	12 cases	9 cases	9 cases
Total	16 cases	11 cases	13 cases
Average	40 per cent	27½ per cent	32½ per cent
Adult and Larval			
Total	46 cases	25 cases	63 cases
Average	34 per cent	19 per cent	47 per cent

The cytoplasmic area was greater than the nuclear in 86 per cent of all cases, the percentage of exceptions being considerably less in the larval than in the adult animals.

TABLE 2

SPECIES	OLD	NEW
Adult		
Fundulus heteroclitus tail	No exceptions	No exceptions
Diemyctylus tail	12 exceptions	9 exceptions
Diemyctylus arm	2 exceptions	7 exceptions
Total	15 per cent	17 per cent
Larval		
Rana sylvatica tail	No exceptions	1 exception
Amblystoma punctatum tail	5 exceptions	2 exceptions
Total	12½ per cent	7½ per cent

Total number of tracings of epithelium.....268
Total number of exceptions..... 38

(c) In a small majority of the cases, 57½ per cent, the number of parts of cytoplasm to one part of nuclear material was found to be greater in the old tissue than in the new. Table 3 shows the summary of the ratios as found for each pair of tracings, the numbers representing X in the ratio

TABLE 3

Summary of tables of areas of cytoplasm to areas of nuclei

NUMBER OF SECTIONS STUDIED	AVERAGE OF PARTS OF CYTOPLASM TO 1 PART NUCLEUS		
	Old	New	
Adult			
10	6.041	6.533	Fundulus heteroclitus tail
64	1.381	1.302	Diemyctylus tail
20	1.428	1.204	Diemyctylus arm
Larval			
10	2.694	2.266	Rana sylvatica tail
30	1.808	1.628	Amblystoma punctatum tail
Total Average.	2.585	2.587	

Area of nucleus : Area of cytoplasm : : 1 : X.

Table 4 shows the percentage of cases for each species in which there is more cytoplasmic material in the old than in the new, as compared with the same amount of nuclear material:

TABLE 4

Adult	*per cent*
Fundulus heteroclitus tail	30
Diemyctylus tail	55
Diemyctylus arm	75
Average	53⅓

TABLE 4—Continued.

	Larval
Rana sylvatica tail	70
Amblystoma punctatum tail	57
Average	63½
Total Average	57½

Summary: From the total number of 536 tracings in this experiment the following conclusions may be drawn: (a) In the mesenchyme of both larval and adult animals and in the epithelium of the larval animals the nuclei are larger in the old tissue than in the new; but in the epithelium of the adult animals the nuclei are larger in the new tissue. (b) In 86 per cent of the tracings of epithelium the cytoplasmic area is greater than the nuclear; the larval tissues show fewer exceptions to this rule than the adult. (c) The amount of cytoplasmic material as compared with nuclear is found to be slightly greater in the old than in the new tissue in 57½ per cent of the cases.

In general then it would seem that in the epithelium of the larval animal the whole cell in the old tissue is larger than the cell in the regenerating tissue, the cytoplasm, however, being larger in greater proportion than the nucleus. In the epithelium of the adult animal the nucleus of the old cell is smaller than that of the cell in the regenerating tissue but the amount of cytoplasm is greater per nuclear area.

11. An attempted analysis of growth. Montrose T. Burrows, Anatomical Laboratory, Cornell University Medical College, New York City.

The study of the growth of different tissues during their development, the study of regeneration, the study of animal behavior, as well as the study of cancer and the effect of physical and chemical conditions on growth and development has shown that the environment is very important in development. Further these studies have given evidence to show that the various forms of cell activity are dependent upon conditions in the environment aside from food and oxygen. Little is known, however, as to the nature of the changes brought about in the cells through this effect of environment nor has sufficient evidence been given to any one theory to cause its general acceptance.

Two years ago I made the observation that growth, division, migratory movements, rhythmical contractions and latency could be observed in heart muscle cells migrating from the same piece of tissue into the same media and in a more recent study I have found that each of these activities is associated with a particular environment. These environmental differences consisted not only in differences in the chemical composition of the medium brought about by the active cell metabolism but also in differences in the mechanical support given these cells, which in plasma clots was altered not only by the concentration and nature of the substances coming from the tissue fragment but also by the shape of the clot and the support given to the clot and the tissue

fragment. The facts in this last statement I have been able to show by direct experiment, as well as to show further that the conditions of the particular environment were necessary for the particular activity shown by the cell. By altering the environment of a cell showing one activity this cell showed the form and activity of one occupying this new environment.

In the paper to be presented before the society I wish not only to describe these observations in greater detail but to give evidence to show that the movement of tissue cells may be interpreted in terms of surface tension. In the same manner I have been able to find a relation between surface tension changes and growth and evidence to show that the organization peculiar to the contracting cells may be interpreted by similar changes.

12. Observations of the lymph-flow and the associated morphological changes in the early superficial lymphatics of chick embryos. Eleanor Linton Clark, Anatomical Laboratory of the University of Missouri.

The present investigation is concerned with a few of the physiological and morphological changes which take place in the developing lymphatic system, after its first appearance. Early superficial lymphatics were studied in living chicks and experiments performed to test the direction and character of the lymph-flow at various stages. The same embryos were then injected and the extent and character of the lymphatic system studied in cleared specimens. Thus an attempt has been made to correlate the structure and function of the early lymphatic system, and to determine, if possible, some of the factors which regulate a few of the phases of its development.

Eggs are opened in a warm chamber, left at incubator temperature, in a manner which has already been described. Under the binocular microscope the lymph circulation is tested by injecting a few India ink granules directly into a lymphatic capillary or duct. The fine glass canula is withdrawn carefully, so as to prevent leakage and the movement of the granules is observed through the binocular microscope. After testing the direction and character of the lymph-flow in various regions, the lymphatic system is injected and the embryo cleared by the Spalteholz method. Chicks of $5\frac{1}{2}$ to 9 days were studied in this manner.

(1) In its primary condition (in chicks of approximately 5 to 6 days) the superficial lymphatic system is a rapidly growing, richly anastomosing plexus. A lymphatic plexus gradually extends posteriorly, from its venous connections in the neck, through the axillary region and down the side. At the same time another plexus is extending anteriorly from the coccygeal veins in the tail. It spreads out over the pelvis and eventually the two plexuses meet and anastomose over the hip. During this period of rapid extension there is no circulation in the superficial lymphatics. The side pressure in the veins with which the lymphatics connect, is higher than the pressure in the lymphatics and con-

sequently blood is continually forced out into the extending lymphatic plexus. The plexus covers a wide area and is irregular and indifferent in character.

(2) The next period in the developing superficial lymphatics is characterized by the beginning of lymph-flow, accompanied by the differentiation of definite ducts or channels in the irregular primary plexus. The flow starts in the side plexus and follows a definite path anteriorly, through the axillary region and the deep plexus, into the veins near the duct of Cuvier. Somewhat later the circulation over the pelvis begins. The flow in this region is instigated by the first pulsations of the lymph heart (still in the form of a plexus). In the earliest stage of circulation the granules move slowly and follow a narrow winding path. Injections show that the first channels are small and somewhat tortuous but quite distinct from the surrounding plexus. On this stage the blood is gradually washed out of the lymphatic system: first from the side region and later from the lymphatics of the pelvis.

(3) The development of the superficial lymphatics in chicks of 7 to 8 days is characterized by increased pressure in the lymphatics, stronger pulsations of the lymph heart, a more rapid lymph-flow and associated with this, the formation of new channels in the lymphatic plexus and the enlargement of those already formed. The exact position of a channel is not predetermined, since variations in the number and position of the main ducts are frequent at all stages.

(4) In chicks of 8 to 9 days, the pressure in the superficial lymphatics is very high. The great increase in the flow of lymph from the allantois and from the deep body lymphatics appears to interfere with the outlet of the fluid from the superficial lymphatics. At this stage the flow is rapid in certain portions of the superficial lymphatic system and very sluggish in others. Injections show that ducts or channels are present in the former regions and large sacs or lakes in the latter. The sacs always occur at a point where there are two conflicting pressures. Because of the looseness of the subcutaneous tissue at this stage, the lymphatic system encounters very little resistance from without and so expands in response to the increased pressure within the lymphatics. The sacs may be formed by the enlargement of two or more neighboring ducts and the breaking down of the walls between them, or by the enlargement of a single duct. At this stage the lymph heart first assumes the form of a sac. Its muscular walls offer an obstacle to its distension and hence it remains much smaller than some of the other sacs or reservoirs. Except for the development of muscles in its wall, the lymph heart does not differ in its mode of formation from other portions of the early lymphatic system.

In addition to the differences in pressure at various stages, the flow of lymph is influenced and altered by (a) the movements of the embryo, (b) the beating of the lymph heart, (c) changes in the blood circulation (d) development of valves at the entrance to the veins, (e) the formation of new lymphatic capillaries and ducts, and (f) by the shifting of the relationship of various organs.

In chicks older than 9 days the increased thickness of the skin and the development of feathers prevented further observation of the circulation of granules in the superficial lymphatics.

13. Studies of the growth of blood vessels, by observation of living tadpoles and by experiments on chick embryos. ELIOT R. CLARK, University of Missouri, Anatomical Department.

There are two views each claiming the support of active workers as to the mode of development of the main arteries and veins. According to one view, which has been largely developed by Hochstetter and recently championed by his pupil Elze, the main arteries and veins develop in definite predetermined places, and the growth of each represents merely the steady extension of a single vessel along its inherited path. The second view, which has been mainly developed by Thoma, and which has found support recently in the works of E. Müller, C. G. Sabin, H. Rabl, Mall and Evans, is that each artery and vein is preceded by an indifferent capillary plexus, any part of which is capable of developing into artery or vein, and that the selection of one or another capillary depends upon mechanical conditions inside and outside the capillary.

The studies here presented are in favor of the second view. The observations on which this view has rested have consisted of studies made by injection or reconstruction, of vessels in a selected region in embryos of different ages. In the present study the development was watched in its various stages in the same embryo. The region selected was the transparent fin expansion of the tail of the frog larva. Drawings were made while the tadpole was immobilized by chloretone, with the aid of an apparatus previously described. By alternating the periods of observation with periods in which the tadpole was returned to fresh water, it was possible to make many successive observations on the same animal, as it increased in size. The records made consisted not only of camera lucida drawings of the vessels, with records as to direction of flow, but also notes as to the comparative rate and amount of flow in each.

It was found that arterioles and venules develop from an indifferent capillary plexus, in which, at any stage, it is impossible to predict which capillary will be incorporated as a part of the advancing arteriole or venule. Thus a vessel which, at one stage, is the main channel between artery and vein, may in later stages either remain the same size, or may even become solid, and disappear by retraction of the endothelium. The factor which determines the selection of a capillary as part of the developing arteriole or venule is the relation in which it is placed with reference to the new capillaries which develop more peripherally. If favorably placed the flow of blood through it increases and its diameter increases, until it becomes a part of the arteriole or venule.

That the development of the main vessels is due to favoring mechanical factors, and not to heredity, is indicated also by the results of experiments on chick embryos. These consisted of the removal of the an-

terior cardinal vein of one side in chicks approximately two days old. This was accomplished by injecting into the vessel Berlin blue—which clumps on contact with the blood and sticks to the endothelium—and removing with forceps and needle the vein and surrounding tissue. The ear vesicle was removed along with the vein as the vein passes under it. In all the chicks in which the operation was successful, examined three or four days after the operation, there was found a well developed vein in the place usually occupied by the internal jugular. In one case this vein was larger than the vein on the unoperated side; in most cases it was slightly smaller, but in all cases it was well developed.

This would seem to show that there exist in the side of the neck mechanical conditions favoring the development of a large vein—since, after the normal vein had been removed, its place was taken by another, which could in no way be considered as inherited.

14. Salient features of the medulla oblongata of Amblystoma embryos of definite physiological stages in development. GEORGE E. COGHILL.

In the stage of development designated by me (Jour. Comp. Neur., vol. 24, p. 163) as non-motile, root fibers of the trigeminal and lateral line ganglia of Amblystoma enter the medulla oblongata. In the early flexure stage the descending trigeminal tract extends to the auditory region and there is a perceptible ascending division of the root; while, in the coiled-reaction stage, the trigeminal tract becomes continuous with the spinal sensory tract, which is composed of fibers from the Rohon-Beard cells. Dorsally of the trigeminal tract in the auditory region of the coiled-reaction stage are recognized the auditory root bundle, the fasciculus communis (solitarius) and two lateral line root bundles, the fasciculus communis laying between the auditory and lateral line root bundles. In the early swimming stage the lateral line root bundles of the seventh and tenth nerves overlap and the fasciculus communis has almost if not quite become continuous with the visceral sensory root bundle of the ninth and tenth nerves. There are no longitudinal association bundles corresponding to tracts *a* and *b* of Herrick (Jour. Comp. Neur., vol. 24, no. 4).

Very large tangential neurones are arranged along the mesial aspect of the root bundles as if functionally related to all of them in common, though smaller cells, located farther dorsad, appear to be especially related to the lateral line components. The axones of the large tangential cells pass mesially of the latero-ventral motor tract to the ventral commissure. This motor tract is the only longitudinal tract in the ventral part of the medulla oblongata until about the time swimming begins, when there appears a suggestion of a slightly more dorsal bundle, presumably the bulbo-spinal tract.

The functional significance of the sensory centers of the medulla oblongata in Amblystoma of this period can not be judged by the degree of development of the sensory root bundles alone, for these are developed entirely out of proportion to the corresponding peripheral nerves,

this being particularly true of the visceral sensory system. Also, experiments show that, in the earlier periods under consideration, the sensibility of the preauditory region to tactile stimulation is much lower than is that of the rostral portion of the trunk.

15. On the development of the lymphatics in the lungs of the pig. R. S. Cunningham, Anatomical Laboratory, Johns Hopkins University.

The lymphatics of the lungs are derived from three sources, the right and left thoracic ducts and the retroperitoneal sac.

In embryos 2.6 to 3 cm. long vessels bud off from the thoracic duct and grow across to the lateral wall of the trachea and form there a plexus that gradually extends over the ventral surface of the trachea and especially down over the bifurcation. From this plexus vessels pass into both lungs and into the pleura.

The right thoracic duct divides, in embryo 2.5 to 2.6 cm., one branch passes to the heart while the other breaks up to form a plexus on the right lateral wall of the trachea; some vessels from this plexus pass down into the hilum of the right lung and others anastomose with the plexus that extends up over the trachea from the other side. The development of the lymphatics within the lung depends upon the division of the vessels into two groups—those accompanying the veins and those accompanying the bronchi and arteries.

Each of the principal branches of the pulmonary vein is accompanied by a group of lymphatic vessels that anastomose freely with the plexus around the adjacent bronchus. These lymphatics grow more rapidly than those associated with the bronchi, and, after following the veins almost to the capillary bed, they pass to the pleura. In the early stages the terminal veins lie about midway between the adjacent bronchi and in this plane a sheet of lymphatic vessels develops from the vessels accompanying the vein and passes to the pleura, marking out the boundaries of the distribution of each bronchus. The first vessels to reach the pleura thus follow the veins, and they anastomose with the vessels that grow to the pleura from the hilum. These vessels reach the pleura when the embryo is about 3.6 cm. long. The bronchial vessels grow more slowly and at first are only to be found around the larger bronchi. As these structures multiply and the lung increases in size the lymphatics accompanying the main bronchi send vessels to the smaller ones, these vessels form a plexus around each bronchus—so that the bronchial tree is surrounded by a continuous series of branching tubes made up of lymphatic vessels. From every point of division of the bronchi lymphatics pass to join those following the veins, and those around the terminal bronchus leave it, near where it ends in the primitive atria, and join those of the veins, septa, or—more rarely—those of the pleura. Lymphatics also arise from the retroperitoneal sac and grow up posterior to the stomach and the diaphragm to enter the lower pole of the lower lobe of the lung. These vessels form a plexus on the median surface of the lower lobe and send branches both to the other surfaces of the pleura of the lower lobe and

into the lung along the veins, where plexuses develop similar to those above and soon the two groups anastomose (embryos 3.9 to 4.1 cm. long).

The further development consists in the multiplication of the plexuses on the bronchi and blood vessels, following the further development of these structures. As the lung increases in volume the larger veins become more closely approximated to the bronchi and only the terminal ones are separated from them, these lie in the periphery of the lobule. Thus the vessels around the veins and bronchi become closely associated, except those accompanying the terminal branches where the veins still lie in the connective tissue septa. These septa develop along the course marked out by the sheets of lymphatic vessels.

The common plexus surrounding the artery and bronchus becomes separated into two plexuses, incident to the increase in the size of the artery, they continue to have many anastomoses however. The vessels of the pleura mark out the early connective tissue septa, but later there develops a fine meshed plexus between these larger divisions, this plexus is not connected with the deep lymphatics. The valves begin to form in embryos about 6 cm. long and practically all point away from the pleura, so that the pleura is drained separately from the remainder of the lung.

In the adult there are lymphatic vessels accompanying the bronchi, the arteries, and the veins—these anastomose freely. There are also vessels in the connective tissue septa that drain chiefly into those around the veins and to some extent into those around the bronchi, near the point where the vein separates from the other structures to take its peripheral position in the lobule. All the deep vessels, together with most of the pleural vessels, drain into large trunks that end in the nodes at the hilum, but the lower half of the pleura of the lower lobe drains by a group of 4 to 6 vessels to the preaortic nodes that develop from the cephalic portion of the retroperitoneal sac. These vessels pass down through the ligament that connects the lower and median surface of the lower lobe with the tissue surrounding the aorta.

16. The morphology of the mammalian seminiferous tubule. George M. Curtis, Anatomical Laboratory, Vanderbilt University Medical School.

The problems here considered may be divided into two phases: (1), dealing mainly with the purely morphologic aspects of the tubule and (2), considering more the relation of the process of spermatogenesis to the tubule.

1. The seminiferous tubule: (a) Blind ends. Since their description and delineation by J. Müller ('30), in the testis of the squirrel, the presence of blind ends in the course of the seminiferous tubules has been repeatedly affirmed and denied. In this work as thus far completed none of these structures have been disclosed. This statement is based upon the following evidence:

Adult albino mouse: Two complete tubules.

One isolated and reconstructed graphically and in wax.
One isolated and reconstructed graphically.
Adult rabbit: Six complete and one incomplete, tubules and tubule complexes.
Five isolated by teasing by Huber (Huber and Curtis '13).
One isolated and reconstructed graphically.
One incomplete complex isolated and partially reconstructed graphically and in wax.
Three-week dog: Two complete tubules.
Both isolated and reconstructed in wax.

From the above eleven tubules and the careful study of the material necessary to isolate them it is concluded that blind ends are not present in these three forms.

(b) Ampullae. These structures described and figured by Sappey ('88) have not been met with in any of the three above forms.

(c) Lobules. These are present in the albino mouse structurally as evidenced by the tubule modelled and by the tubule graphically reconstructed. However, no apparent lobulation is visible in examining the sections. In the rabbit and dog lobules are visible with the naked eye, each lobule being found to contain the coils of a portion of a single tubule or tubule complex.

(d) Branches and anastomoses. These were found to be infrequent in the mouse testis, more frequent in the dog and most frequent in the rabbit.

(e) Embryonic ends. In the mouse tubule, between the cessation of the active process of spermatogenesis and the flattened epithelium of the tubules rectus, was found a region where the tubule retained its embryonic structure, disclosing the sexual and sustentacular cells around an irregular lumen. It suggests itself that this may be a possible region of reserve to be used in growth or regeneration.

2. The spermatogenic wave: Especially through the work of v. Ebner it has been shown that the development of mammalian spermatozoa proceeds in a wave-like process along the course of the seminiferous tubules. v. Ebner ('88) states that in the rat these waves ascend from the rete and vary in length from 25 mm. to 38 mm. averaging 32 mm. Benda ('87) intimates their variability.

A study of these waves has been made in the two complete seminiferous tubules of the adult mouse, and in one complete and in a portion of an incomplete tubule complex in the rabbit. In determining the relations between wave and tubule it first became necessary to arbitrarily choose a series of eight successive stages of spermatogenesis. These were obtained from v. Ebner's figures and a study of the series. The above tubules were then reconstructed graphically, their loops corresponding to the numbered tubule sections in the serial drawings. By observing the stages of spermatogenesis present in each tubule section at definite intervals and applying them to their proper loop in the graphic reconstruction, the continuity of the stages was determined.

By comparing and numbering alike all the loops of the serial drawings,

graphic reconstruction and model, the relations of the waves to the model were determined and their actual lengths computed. Their succession was shown by plotting the successive stages occurring along the course of the tubules. By this method the following results have been obtained.

(a) Wave length. From a study of seven waves in the mouse this was computed as averaging 1.83 cm. From a study of one complete wave and a comparison of eight wave portions the average wave length in the rabbit is estimated 1.4 cm.

(b) Wave variability. In the mouse and rabbit the waves vary in length, direction of course, uniformity, and in that single stages may be out of order in a successive series.

(c) Wave reversibility. In both forms the waves may reverse their direction, often frequently in a single portion of a tubule or tubule complex.

(d) Wave direction. In the mouse the waves of five rete ends all descend from the rete. In the rabbit the waves vary, three waves ascending and two descending from the rete.

The above work was completed under Dr. Huber's direction at the Histological Laboratory of the University of Michigan, and I desire to express here my thanks for his courtesies and assistance.

17. The structural relations of anterior hepatic anteries. C. H. Danforth, Washington University Medical School.

In an earlier paper (Jour. Morph., vol. 23, no. 3, 1912) the writer published a brief description of the anterior hepatic arteries of Polyodon. These vessels, which arise from the same trunk as the posterior coronary arteries, were found to be of constant occurrence and of fairly uniform distribution. They were often equal to, or even more extensive than the posterior (ordinary) hepatic arteries, with which they anastomose.

At the time the above mentioned paper was published, it was supposed that anterior hepatic arteries were peculiar to Polyodon. Further observations, however, have revealed them in several other forms. Their general relations, so far as gross methods reveal, seem to be essentially the same in all cases.

It is now possible to record a few recent observations on the development and finer relations of the anterior hepatic arteries of Polyodon. In this fish the connective tissue about the hepatic veins is unusually extensive. Associated with these veins, as well as with the branches of the portal system, there are considerable accumulations of lymphoid tissue. In general there is a branch of the artery running through each of these lymphoid aggregations. In young fish, less than 100 mm. in length, the thickened connective tissue sheath about the vein is not apparent and in such specimens anterior hepatic arteries have not been detected. At 123 mm. the connective tissue about the veins shows a slight thickening and the arteries may be traced in sections. But even at this stage there is no noticeable accumulation of lymphocytes.

In the adult the ramifications of the artery are usually found associated with the tributaries of the hepatic vein. Nevertheless, they are not confined to the connective tissue about the veins, but branches of considerable size may pass out into the liver parenchyma where they are for the most part surrounded by lymphocytes. Some of their branches may again become associated with veins.

These observations indicate that the anterior hepatic arteries are not to be regarded as of the nature of vasa vasorum in connection with the hepatic veins, but as independent vessels, probably of considerable functional importance.

18. The so-called "endothelioid" cells. Hal Downey.

Pathologic conditions affecting primarily the hematopoietic organs are frequently characterized by the presence of large protoplasmic cells which are usually designated as "epithelioid" or "endothelioid" cells. Such cells are seen in generalized granulomata of the lymph nodes (tubercular lymph nodes, Hodgkin's disease), in Gaucher's disease, Banti's disease, in lymph nodes from typhoid fever patients, etc. Although these cells may show structural variations of considerable degree, most pathologists, especially American pathologists, do not hesitate to group them together under the heading of "endothelioid cells" or 'endothelial leucocytes.' They are given this name, because it is believed that they are derived from the endothelium which is supposed to line the lymph sinuses and cover the strands of reticulum of lymph nodes, or from the "endothelial" lining of the venous sinuses of the spleen, or from that which lines the blood and lymph vessels, primarily the latter.

Mallory calls all these cells 'endothelial leucocytes,' and he believes that they correspond to the so-called "large mononuclear leucocytes" of the circulating blood. Of the latter he says (Principles of Pathologic Histology, p. 21): "They are derived from the endothelial cells lining blood, and to a less extent lymph, vessels by proliferation and desquamation. They also multiply by mitosis after emigration from the vessels into the lesions." In this connection Mallory's idea of the structure of a lymph node is of interest. On page 616 he states: "Next to the cells of the lymphocyte series the most important cells of the lymph nodes are the endothelial cells. They line the blood vessels, the lymph sinuses and the reticulum of the parenchyma. Those lining the sinuses and the reticulum play a much more important part in pathologic conditions than those lining the blood vessels. They may increase greatly in number, desquamate from the walls of the sinuses and from the reticulum and form endothelial leucocytes. As a rule they exhibit marked phagocytic properties for other cells The capsule and trabeculae are composed of fibroblasts, among which are occasional smooth muscle cells. Fibroblasts also form the reticulum in the lymph sinuses and in the parenchyma and strengthen the walls of the vessels."

From this we see that Mallory believes the entire reticulum of a lymph node to be covered by a distinct endothelium which is independ-

ent of the reticular cell, which he describes as fibroblasts. This endothelium not only lines the sinuses but also covers the reticular strands of the parenchyme. Mallory is quoted so extensively because, in the main, his views coincide with those of most American pathologists.

The writer became interested in this problem while working over the lymphoid tissue of a fish (Folia Haem., Bd. 8). Here it was found that the blood and lymph spaces of the lymphoid tissue were not lined by a distinct endothelium, and that cells which might be mistaken for endothelial cells were merely portions of the general reticulum, in many cases with fibrils running through their protoplasm. The reticulum was partly fibrous and partly protoplasmic; where the fibers were present they were always embedded in the protoplasm of the reticular cells. The question was investigated again in connection with a problem on the origin of the lymphocytes in lymph nodes and spleen (Arch. f. mikr. Anat., Bd. 80), and lately in connection with a study of the histology of the spleen and lymph nodes in Gaucher's disease.

The conclusions from this study of the reticulum and its supposed relations to endothelial cells are very different from those of such anatomists as v. Ebner and Stöhr, and the greater number of pathologists. Even with ordinary methods it is evident that the strands of the reticulum are composed of branched, anastomosing cells which are closely associated with the fibers. Nothing can be seen of a continuous epithelial covering. Associated with these strands, especially where the reticulum forms the wall of a sinus, are varying numbers of larger and more rounded protoplasmic cells whose connection with the fibers of the reticulum is not so evident with ordinary methods. Such cells, especially where they project out into the lumen of a sinus, might well be mistaken for hypertrophied endothelial cells. However, the use of any one of the numerous specific stains for reticular fibers (Krause's iodo-iodide of potassium—gold chloride method, the Maresch-Bielschowsky, or the older formula of Mallory's hematoxylin as used by Thomé) shows clearly that these cells are frequently traversed by fibers, and that even the large rounded cells resembling large mononuclear leucocytes are frequently attached to the reticulum and have fibers embedded in their peripheral portions. These latter cells show great phagocytic activity, especially for red corpuscles, and their nuclei are large and indented. If these cells were not attached we would not hesitate to pronounce them as large mononuclear leucocytes. Frequently large numbers of similar cells are seen free in the sinuses and in the meshes of the reticular network. It is no difficult matter to show that they have been derived from the reticulum. These same cells are very numerous in the lymph of the thoracic duct and in the lymph of the lymph vessels beyond the lymph nodes. It therefore seems proven that large mononuclear leucocytes, or at least cells which cannot be distinguished from them morphologically, may be derived from the reticulum of the lymph nodes; in fact it is possible to demonstrate all intermediate stages between ordinary reticular cells and these larger cells. These cells

are frequently seen to be dividing by mitosis both within the lymph nodes and within the thoracic duct. The resulting daughter cells will be smaller cells resembling lymphocytes in structure.

The specific stains for reticular fibers, especially when followed by a good counterstain, show further, that the reticular fibers of the strands within the parenchyme are embedded in the protoplasm of the cells. With these methods it is impossible to see an endothelial covering to these strands. Since there is no endothelium covering the reticular strands or lining the sinuses we are hardly justified in naming the large cells which are cut off from the reticulum 'endothelial leucocytes.' Whether such cells may also be derived from the endothelial cells lining lymph and blood vessels, as claimed by Mallory, still remains to be demonstrated. Theoretically there is nothing against such a view, since numerous investigators, including the writer, have shown that similar cells may be derived from the covering cells of the omentum and serous layers generally. However, Weidenreich and Schott believe that these covering cells are merely flattened surface fibroblasts (see also experiments of W. C. Clarke, Anat. Rec., vol. 8, no. 2, p. 95). If this view is correct these covering fibroblasts would not be very different from the reticulum cells of the lymph nodes.

There is nothing new about the results obtained from this study of normal animals, since Thomé, Weidenreich and others reached the same conclusions. The pathologists Rössle and Yoshida, working with the Maresch-Bielschowsky method, also concluded that it is impossible to distinguish between endothelial cells and cells of the reticulum, and Ferguson, using the same method, found that the fibers of the reticulum are largely embedded in the protoplasm of the reticular cells. This literature, however, is almost unknown to pathologists; consequently statements like those quoted from Mallory are constantly reappearing in the pathological literature, and to some extent in the anatomical literature also (Evans, Anat. Rec., vol. 8, no. 2, p. 101).

Gaucher's disease has already been mentioned as one of those diseases which are characterized by the presence of large numbers of the so-called 'endothelioid' cells in the spleen, lymph nodes, liver, and bone marrow. The writer was fortunate in obtaining some of this material from Dr. F. S. Mandlebaum (Pathologist, Mount Sinai Hospital, New York City). The fixation of the material is unusually good, and so it is ideal material for working out the origin of the 'endothelioid' cells. In this case these cells are large clear cells characterized by the presence of exceedingly fine fibrils in their cytoplasm. American pathologists have claimed that they were derived from the endothelium, especially from that lining the venous sinuses of the spleen, and the lymph sinuses of the lymph nodes. Several German pathologists have suspected that the reticulum was concerned in the formation of these cells, but none of them were able to prove this positively, as they could not find the necessary intermediate stages between reticular cells and the large cells of the disease. Mandlebaum's material,

however, shows the early stages in the disease, and it is not difficult to find all of the necessary intermediate stages between the characteristic cells of the disease and the cells of the reticulum, as I hope to be able to prove with the demonstrations. In the liver, in the walls of the vessels, etc., they seem to be derived from fibroblasts. From this material it was impossible to prove the origin of these cells from the 'endothelium' of the venous sinuses of the spleen. However, if such were the case it would in no way invalidate the above findings, as it is now generally conceded by anatomists that this 'endothelium' is merely a specially modified portion of the reticulum.

In Hodgkin's disease we again have cells which have been called 'endothelioid' cells. They are very different in character from the large cells seen in Gaucher's disease, nevertheless, their origin from the reticulum of the lymph nodes is easily demonstrated. Reticular fibers may be seen penetrating their protoplasm, and the same is true of the Gaucher cells while they are still attached to the reticulum.

These facts, and the results obtained from a study of normal lymph nodes, show that the large cells which are characteristic of many pathologic processes, and which are numerous in the sinuses of normal lymph nodes, in the lymph of the thoracic duct, etc., are in most cases not derived from endothelial cells. There is, therefore, no reason for naming them 'endothelial leucocytes' or 'endothelioid' cells. In most cases they could be designated as "reticular" cells. However, this would not do for a general term, because Dominici, Weidenreich and Downey among others have shown that large mononuclear leucocytes may also be derived from lymphocytes. The intermediate stages in this process are shown in one of the lantern slides. One of the slides will also show that the reverse may be true, i.e., that lymphocytes may be derived from large mononuclear leucocytes.

Those investigators (Goldmann, Evans and Schulemann, Aschoff, Kiyono, etc.) who have recently been engaged in the study of the results of vital staining by means of lithium carmine and the dyes belonging to the benzidine group will not agree with the view of the relationships between cells of the reticulum and large mononuclear leucocytes and lymphocytes expressed above. However, it must be remembered that they have not yet succeeded in showing that the cells which are able to store the dyes in the form of granules (a process related to phagocytosis—Evans and Schulemann) are *genetically* different from those which do not take up the dye. Their results are equally well explained if we assume that those lymphoid cells which are located in the tissues or which have recently been cut off from the reticulum are in a condition which is especially favorable for phagocytosis, a fact which was known long before the modern investigations with vital staining were begun. We know that reticular cells, while they are still attached to the reticulum, show special phagocytic activity, and that this activity may be increased after the cells have separated from the reticulum. This can easily be proven by an examination of the large reticular cells in the sinuses of any lymph node

which contains free red corpuscles. In the lymph of the thoracic duct or in the peritoneal fluid the phagocytic activity of these cells is still very pronounced, but it is greatly diminished as soon as they reach the blood stream. In the tissues the phagocytic activity is again very pronounced. The function, and to some extent the morphological appearance, of a lymphoid or 'endothelioid' cell depends, therefore, on the conditions under which it finds itself. It is not necessary to assume the existence of a special line of 'histiocytes' which differ genetically from the other lymphoid cells.

19. On the anlage of the bulbo-urethral and major vestibular glands in the human embryo. (Lantern). Arnold H. Eggerth, Department of Anatomy, University of Michigan. (Presented by G. Carl Huber.)

For this investigation, the urogenital systems of four human embryos of critical ages from the collection of Dr. Huber were reconstructed. In each of the models, the urogenital sinus presents three pair of symmetrically placed lateral epithelial folds. The cephalic ends of the middle of these lateral folds bear short epithelial buds, the anlagen of the bulbo-urethral and major vestibular glands. The measurements given are for crown breech length. The model of a 32 mm. female embryo presents a short epithelial bud, 15 μ in length, on the left side only. The model of a male embryo of 30 mm. presents gland buds on both sides, whose respective lengths are 50 μ and 60 μ. A female embryo of 45 mm. shows gland buds having a length of 120 μ and 150 μ, and a female embryo of 60 mm. presents gland buds with terminal branching, having a length of 200 μ and 240 μ. The relative positions for the gland anlagen in both male and female embryos for the varying ages as reconstructed, is essentially the same.

20. The cell clusters in the dorsal aorta of the pig embryo. V E. Emmel, Department of Anatomy, Washington University Medical School.

In the course of a study of hematogenesis in several regions of the mammalian vascular system, the following observations were made on the dorsal aorta of the pig embryo. The material studied consisted of about seventeen embryos varying from 6 to 25 mm. in length, together with several mouse and rabbit embryos. In the dorsal aorta of the 6 to 15 mm. specimens there occur rounded cell masses or clusters, the cytological characteristics of which identify the component cells as belonging to the mesamoeboids of Minot or the primitive lymphocytes of Maximow. Their constant occurrence at certain stages of development, their evident more or less firm attachment to the vascular surface, and their restriction, apparently without exception, to the ventral wall of the aorta, appear to necessitate relegating to these clusters a significance greater than that of agglutinated cell masses merely incidentally resting upon the aortic wall. The absence, in many cases, of evident endothelial continuity at the basal regions of these clusters, the transitional cytological characteristics from the

basal to the more peripheral cells, the changes in form and increase in number and size of the adjacent endothelial nuclei, together with the frequent occurrence of mitotic figures within the masses, is evidence strongly indicative of their active proliferation and origin *in situ* from the aortic endothelium. At the 15 mm. stage the clusters have become greatly reduced in number and are no longer to be observed in the 25 mm. embryo. During this 6 to 20 mm. period of development, there occurs in the ventral region of the aorta, in contrast to the dorsal region, an extensive degeneration of the medial and lateral intersegmental aortic arteries and a remarkable 'caudal wandering' of the coeliac and mesenteric arteries upon the aortic wall. The simultaneous appearance of these phenomena in the ontogeny of the embryo and the morphological interrelationships of the structures under consideration in the ventral aortic wall are of such a character as to be suggestive of some significant correlation between the formation of these clusters and the development of the permanent visceral arteries of the adult.

21. Feeding experiments on rats. J. F. GUDERNATSCH, Department of Anatomy, Cornell University Medical College, New York City.

Based upon the results obtained by feeding the internally secreting glands to amphibians, these experiments are being continued, this time with mammals. The glands are given to white rats in stated portions, at regular intervals. A preliminary account of some observations on the thyroid-treated animals may here be given.

Beef thyroid was fed in portions small enough to keep the animals in fairly good health; 1 gram a week was given, in some experiments 1 gram in 5 days. The application of even so small doses of thyroid sometimes produced slight symptoms of hyperthyroidism; however, the animals kept well enough to have offspring.

The following enumeration gives the records of 8 successful matings; in the first 4 cases the offspring are still living, while in the remaining 4 the young died, at stated dates.

Case I: ♂ *t* × ♀ *t.*[1] (a) While under treatment the father was bred to 3 treated ♀; no result. (b) After discontinuation of the thyroid treatment the father was bred to a non-treated ♀; 10 young were born after 26 days, all so frail that they died within a week. (c) One month after thyroid treatment of the father and immediately after thyroid treatment of the mother the two were mated; 3 young were born 54 days after the father and 29 days after the mother had received their last dose of thyroid. The young are much smaller than the normal rats of equal age.

Case II. ♂ *t* × ♀ *n.* After discontinuation of the thyroid treatment the father was bred to the non-treated mother; 4 young were born 30 days later; 2 died very soon, 2 undersized ones are living.

Case III: ♀ *t* × ♂ *n.* (a) While under treatment the mother was

[1] *t* = treated; *n* = normal.

bred to a treated ♂; no result. (b) Immediately after thyroid treatment the mother was bred to the non-treated father; 6 young were born 113 days later; 4 undersized ones are living. The mother required 3 months to recover from the thyroid influence.

Case IV: ♂ *t* × ♀ *n.* (a) While under treatment the father was bred to a treated ♀; no result. (b) The normal mother was bred to 2 normal ♂; 2 litters. (c) After discontinuation of thyroid treatment the father was bred to the normal mother; 3 young were born 117 days later; they are undersized. The father required 3 months to recover from the thyroid influence.

Case V: ♂ *t* × ♀ *n.* (a) While under treatment the father was bred to a non-treated ♀; no result. (b) While under treatment the father was bred to the non-treated mother; 5 young were born 24 days later; 1 died 9 days old, 4 very frail and undersized lived about 7 months. When 3 months old they weighed 57, 58, 60 and 66 grams respectively; (65 to 70 grams is the average weight of a rat about 60 days old).

Case VI: ♂ *t* × ♀ *t.* (a) While under treatment the father was bred to the treated mother; no result. (b) While under treatment the father was bred to a non-treated ♀; 5 young (see Case V). (c) The treated father and treated mother (under a) were again mated; thyroid treatment ceased 31 days later; 7 young were born 81 days later; very frail and undersized; lived 2 months. The parents required 2 months to recover from the thyroid influence.

Case VII: ♂ *t* × ♀ *n.* (a) While under treatment the father was bred to 3 treated ♀; no result. (b) After discontinuation of the thyroid treatment the father was mated to the non-treated mother. Ten young were born 26 days later; all died within 2 weeks.

Case VIII: ♀ *t* × ♀ *n.* (a) The non-treated father was bred to 2 non-treated ♀; 2 litters. (b) Before thyroid treatment the mother was bred to a non-treated ♂; 1 litter. (c) While under treatment the mother was bred to a treated ♂; no result. (d) After discontinuation of the thyroid treatment the mother was bred to the non-treated father; 5 young were born 151 days later; all died within 5 days. The mother required 4 months to recover from the thyroid influence.

The history of these cases shows that the feeding of thyroid to rats greatly interferes with their breeding qualities. Twenty-four matings, in which both parents were treated, resulted in failure, 2 in which the female alone had been treated and 4 in which the female alone received thyroid food, in all 30 matings. Yet out of these 14 males 7 had been tested and given offspring previously to the treatment, and out of the 16 females 9 had been tested and were found fertile.

Table 1 gives the enumeration of several matings, which will show that pregnancy did not set in until several weeks after the discontinuation of the thyroid treatment, except when the female was non-treated. The number of days is given that elapsed between the placing together of the parents and the birth of a live litter (or death of the female). The gestation period of the rat is from 21 to 24 days.

Thus under no circumstances will pregnancy set in during the thyroid treatment (cases 3 to 7, 15); after discontinuation of the thyroid treatment, the animals usually required several weeks to recover from the thyroid influence (cases 11 to 18).

TABLE 1

(1) ♀ dies after 2 days		Thyroid given after mating
(2) ♀ dies after 2 days		
(3) ♀ dies after 7 days	No pregnancy	
(4) ♀ dies after 19 days		
(5) ♀ dies after 21 days		
(6) ♀ dies after 21 days		
(7) ♀ dies after 38 days		
(8) ♀ normal; young born after 24 days; all die early		No thyroid given after mating
(9) ♀ normal; young born after 26 days; all die within two weeks		
(10) ♀ normal; young born after 28 days; 2 die within a week		
(11) ♀ dies after 57 days; 6 fetuses		
(12) ♀ dies after 67 days; 7 fetuses		
(13) ♀ dies after 107 days; pregnant		
(14) ♀ dies after 112 days; pregnant		
(15) young born after 110 days, 87 days after thyroid treatment		Thyroid feeding continued for 33 days after mating.
(16) ♂ normal; young born after 113 days		No thyroid given after mating
(17) ♀ normal; young born after 117 days		
(18) ♂ normal; young born after 151 days		

Only one mating of both parents previously treated gave offspring after 29 days. However, the treatment of the father had ceased one month before the mating time.

The feeding to rats of fresh thyroid tissue shows its effect in three different ways:

1. When the dose is too large, all the well-known symptoms of hyperthyroidization become evident, viz.: emaciation, diarrhoea, muscular weakness and finally cachexia leading to death. The hair becomes yellowish, stands erect, sometimes falls out in patches, in short the entire coat looks ragged.

2. When the dose is so regulated, as to keep the animals in approximately good health—the fur will always become shabby—then the animals do not breed. *Not one mating of both parents treated*, after the animals had been placed together, *gave any result.* Pregnancy was always delayed, since fertilization did not occur until several weeks after the application of the thyroid had been discontinued.

3. Did pregnancy finally occur, it resulted (a) in abortus; (b) the young died soon after birth; (c) in very late pregnancies, the young show a diminished tendency to grow. Although they are not especially frail, they keep in relative size behind the young of normally fed rats.

22. The development of reflex mechanisms in Amblystoma. C. Judson Herrick and George E. Coghill.

The results of neurological studies of Amblystoma by the authors and others now afford a tolerably sound basis for the interpretation of some features of the mechanism of functional differentiation of the central nervous system of the individual and may also contribute something to the knowledge of the factors involved in the phylogenetic differentiation of the nervous system.

Most of the observations on the nervous system of Amblystoma and other urodeles from which these conclusions have been deduced are recorded in the following papers:

Coghill, George E. 1902 The cranial nerves of Amblystoma tigrinum. Jour. Comp. Neur., vol. 12, pp. 205–289.

1909 The reaction to tactile stimuli and the development of the swimming movement in embryos of Diemyctylus torosus Eschscholtz. Jour. Comp. Neur., vol. 19, pp. 83–105.

1913 The primary ventral roots and somatic motor column of Amblystoma. Jour. Comp. Neur., vol. 23, pp. 121–143.

1914 Correlated anatomical and physiological studies of the growth of the nervous system of Amphibia. I. The afferent system of the trunk of Amblystoma. Jour. Comp. Neur., vol. 24, pp. 161–233.

Herrick, C. Judson. 1914 The medulla oblongata of larval Amblystoma. Jour. Comp. Neur., vol. 24, pp. 343–427.

The swimming reflex. The reflex mechanism essential to swimming in Amblystoma embryos of the youngest age in which this reflex is possible consists of three groups of neurones: (1) sensory peripheral neurones lying within the spinal cord (the transitory Rohon-Beard cells) which send their dendrites to the skin and myotomes, while their axones ascend in a dorso-lateral sensory tract of the cord; (2) commissural neurones which pass from the sensory cells of one side to the motor cells of the other through the ventral commissure; the decussation of these fibers occurring only in the upper spinal cord and lower medulla oblongata; (3) motor cells, which form a descending, ventro-lateral motor tract and innervate the myotomes by means of collaterals. It should be noted particularly that all responses of such embryos are 'total reactions,' and of the same sort regardless of the place and kind of excitation; that the peripheral sensory fibers are not specific with reference to exteroceptive and proprioceptive stimuli; that the sensory and motor peripheral neurones are not differentiated away from central neurones of longitudinal columns, and that the first central paths to appear are long and made up of chains of numerous relatively short neurones.

Spinal reflexes in half-grown larvae. The spinal ganglion cells and ventral horn cells are at this age fully matured, and crossed as well as uncrossed reflexes have become possible at all levels in the spinal cord. Both correlation neurones and ventral horn cells send dendrites into all parts of the cross section of the white substance, some even

crossing in the ventral commissure. The responses are still in large measure simple and 'total reactions,' but they are brought under the influence of a much greater variety of excitations than are those of young embryos, in consequence of the introduction of longitudinal tracts that are actuated by special sense organs, especially those of the head.

The mammalian spinal cord. Here the correlation neurones are organized into an elaborate system of distinct reflex circuits, and there is a corresponding specialization and refinement of motor functions.

The medulla oblongata of larval Amblystoma. Each physiological type of end organ has its own distinct ganglion or ganglia and nerve roots. Each root fiber from the several types of end organs, immediately upon entering the medulla, divides into ascending and descending branches which pass upward and downward for practically the entire length of the medulla oblongata. These bundles of root fibers constitute nearly all of the substantia alba of the dorsal half of the medulla, with the exception of two large, longitudinal correlation paths on either side. The ventral half of the white substance contains the motor roots and numerous long correlation tracts. In contrast with the sharp physiological differentiation of the sensory neurones of the first order, those of the second order are not functionally specific, for the dendrites of any one of them may establish synaptic relations with several or all of the peripheral sensory root bundles. The primary sensory centers, therefore, serve, not only as receptive centers, but also as correlation centers.

The medulla oblongata of mammals. The arrangement of the peripheral sensory neurones in the mammalian medulla oblongata is essentially the same as in the amphibian. The sensory neurones of the second order, however, are segregated into definite primary receptive centers, each related specifically to one peripheral system, and the secondary paths leading from these primary centers may be as specific functionally as are the peripheral root bundles themselves. The correlation of these elements into particular reflex systems is effected in centers farther removed from the first sensory neurone of the arc.

Conclusion. It is the prevailing belief that every form of central nervous system has arisen by the concentration of an original diffuse and relatively equipotential peripheral ganglionic plexus. Out of such a primordial nervous matrix there has been a progressive individuation of centers and pathways and a parallel progressive differentiation of specific reflexes away from the primitive type of 'total reaction.' The reflex mechanisms of embryonic and larval Amblystoma are in many respects primitive; and their forms suggest that they represent different stages in this process of individuation of specific reflexes. The 'typical' two-neurone, short circuit connection between dorsal and ventral root fibers is, therefore, not to be regarded as primitive. During such processes of individuation of parts of the nervous system its integrative action has been preserved through the development of correlation centers farther removed from the receptors and effectors. Thus arose such supra-segmental apparatuses as

the cerebellum and cerebral cortex. Finally, we would urge that the factors operating in either the ontogenetic or the phylogenetic differentiation of the functional mechanisms of the brain cannot profitably be investigated without a precise knowledge in each stage investigated of the peripheral relations of each of these functional systems and of the interrelations of the neurones involved at every step in the progress of the nervous impulse from periphery to center and back to the effector organs during the normal course of functional activity.

23. The development of fibrous tissues in peritoneal adhesions. Arthur E. Hertzler, Kansas City, Mo.

The material used in this study was obtained by causing adhesions of intestines by suture or by irritants and by attaching foreign bodies to the mesentery or omentum.

When a suture of adjacent loops of intestine is placed, the space between the guts is filled with an amorphous exudate. In 10 to 30 minutes this exudate coagulates forming fibrinous bands which extend from one gut surface to the other. These bands stain specifically with Weigert and Mallory stains. By comparing series it can be noted that the bands first lose the specificity for Weigert while retaining it for Mallory. This occurs in 24 to 48 hours. In 4 days they no longer accept the Mallory stain for fibrin but do accept the Mallory fibril stain. Bands may be seen which stain in part red and in part blue with the Mallory stain. With picro-fuchsin the same transition attains.

When a disc of foreign material is sewed to the mesentery it is covered at once with an exudate. This coagulates over the entire surface and its conversion into fibrous tissue takes place simultaneously over the entire disc and does not proceed from the edges toward the center.

These fibrin bands may form in the exudate before the advent of cellular elements and the transition above noted may take place without the advent of cells.

Any process which prevents the exudate from coagulating into fibrin prevents wound healing. This is true irrespective of the means employed. Infections produce this effect permanently and peptonization of the animal prevents it temporarily.

If wound healing has been delayed by any means which prevents the formation of fibrin in the primary exudate then healing must await the advent of new exudate. This takes place only when cells find their way into the unfriendly exudate. The formation of fibrous tissue then takes place according to the methods described in the literature. The healing of wounds as described in the literature is in fact healing by second intention.

24. On the development of the digitiform gland in Squalus acanthias. E. R. Hoskins, Institute of Anatomy, University of Minnesota.

The digitiform gland in Aqualus is evidenced first by a slight thickening of the entoderm of the dorso-lateral border of the gut just pos-

terior to the spiral valve. This may be seen in embryos 15 mm. in length, especially in those sectioned longitudinally. The thickening soon pushes laterally to form a hollow bud which turns and grows anteriorly along the gut. The form of the curved portion at the point of emergence from the gut always persists so that in older stages and in the adult this portion which becomes the duct of the gland enters both the intestine and the digitiform gland anteriorly.

In the stage of 28 mm. it may be seen that from the main part of the gland small buds resembling the original form of the gland grow laterally on all sides. These buds become tubules extending laterally and slightly posteriorly. They in turn give rise to secondary tubules which in time form irregular groups opening into the primary tubules. This condition is to be found throughout development, the gland becoming a compound tubular structure, the secondary tubules arising from the primary, close to the main lumen of the gland.

As the gland develops, it carries the mesentery of the intestine with it and is thus supported from the dorsal wall of the body cavity.

The entoderm of the digitiform gland is composed at first of four layers of low columnar or cuboidal cells with elongated nuclei, being similar to the entoderm of the gut from which it develops. As the gland increases in length, the epithelium is gradually reduced to one layer in thickness. Its primary and secondary tubules both arise as structures of an epithelium of one layer of cells. At the points of greatest growth, namely, at the distal ends of the tubules the nuclei are wider and shorter than along the main lumen, often being spherical.

The epithelium lining the main or central lumen later thickens giving us a structure of two layers of columnar cells with rounded nuclei in the full-term fetus and of four layers in the adult.

25. The development of the albino rat, from the end of the first to the tenth day after insemination. G. Carl Huber, Department of Anatomy, University of Michigan.

The material on which this investigation is based was collected while the writer was stationed at The Wistar Institute of Anatomy. For the trustworthiness of the records pertaining to the time of insemination of the female rats used, he is greatly indebted to Dr. J. M. Stotsenburg. The age of the stages as given in this account is reckoned from the time when copulation was first observed, thus from the time of insemination. Carnoy's fluid was used as a fixative; paraffin embedding and staining in hemalum and Congo red was the general procedure. The process of ovulation, maturation and fertilization having been carefully studied by Sobotta and Burckhard, their account carrying the development to the pronuclear stage, my own studies of the development of the albino rat begin with this stage.

The pronuclear stage extends through a relatively long period, perhaps 12 to 15 hours. All ova obtained 24 hours after insemination present the pronuclear stage, this period presenting about the middle of the pronuclear phase. Of the two pronuclei, the female

pronucleus is slightly the larger. The nuclei lie near the centre of the ovum, are distinctly membranated, and do not fuse prior to the formation of the first segmentation spindle. By the end of the first day, the fertilized ova have travelled about one-fourth the length of the oviduct, and are found lying free in its lumen. The formation of the first segmentation spindle, and the first segmentation occur during the early part of the second day after insemination. The resulting 2 cell stage extends for a period of about 24 hours, since 2 cell stages were found in material obtained 1 day, 18 hours to 2 days, 22 hours after insemination. The first two blastomeres are equivalent cells. By the end of the second day after insemination, all the fertilized ova are in the 2 cell stage, having traversed a little over one-half the length of the oviduct. One of the cells of the first two blastomeres divides before the other resulting in a three cell stage; such a stage was obtained 2 days, 19 hours, and 2 days, 22 hours after insemination. The division of the other of the first two blastomeres soon follows, so that by the end of the third day all tubes examined contained ova in the 4 cell stage, they having traversed by this time about $\frac{9}{10}$ of the length of the oviduct.

An 8 cell stage is reached toward the end of the fourth day after insemination (3 days, 17 hours) and by the end of the fourth day, the segmenting ova, in a 12 cell to 16 cell stage, pass from the oviducts to the uterine horns.

It will be observed that beginning with the pronuclear stage, found 24 hours after insemination, there occur three successive segmentations, spaced at intervals of about 18 hours and resulting in 2, 4, and 8 cell stages during transit of the ova through the oviducts. During the fourth segmentation, the ova pass from the oviducts into the uterine horns. Weighings of the water displaced by a series of madels made of early segmentation stages indicate that during the first 4 days of the development of the albino rat there is only very slight increase of the size of the egg mass as against the unsegmented ovum with two pronuclei.

During the early hours of the fifth day after insemination all of the segmenting ova of the albino rat are to be found lying free in the lumen of the uterus, spaced as in later stages of development, the fifth series of segmentations having been completed by this time, the resulting morula mass having an ovoid form and consisting of 24 to 32 cells and measuring approximately 80 μ by 50 μ. During the middle of the fifth day after insemination, the early stages of blastodermic vesicle or blastocele formation may be found. The segmentation cavity or blastocele begins as a single, irregularly crescentic space, arising between cells, and is excentrically placed. The early stages of blastocele formation are thus observed in morula masses composed of 30 to 32 cells, these lying free in the cavity of the uterus. The enlargement of the blastocele, after its anlage, is obtained by a flattening of the border or roof cells; to a lesser extent, by an increase in the number of these cells. By the end of the fifth day after insemination,

all the ova are found in the blastoderm vesicle stage, one pole of each vesicle, designated its floor, consisting of a relatively thick mass of cells; the other pole, its roof, consisting of a single layer of flattened cells bordering and enclosing the segmentation cavity. Cell differentiation into a layer of covering cells, a layer of ectodermal and entodermal cells, such as described by Selenka, is not to be observed at this stage. During the sixth day after insemination, at which time the ova still lie free in the lumen of the uterine horn, the blastodermic vesicles increase in size, partly as a result of further flattening of the roof cells, partly as a result of rearrangement and flattening of the cells constituting the floor of the vesicle, this portion of the vesicle now consisting of about three layers of cells, the innermost layer having differentiated to form the yolk entoderm. By the end of the sixth day the blastodermic vesicle consists of a discoidal area, the germ disc, comprising about $\frac{1}{5}$ to $\frac{1}{6}$ of the wall of the vesicle, and consisting of two to three layers of cells, of which the inner is differentiated to form the yolk entoderm, the remainder of the vesicle wall consisting of a single layer of very much flattened cells.

During the seventh day after insemination, the blastodermic vesicles become definitely oriented in the decidual crypts, the thicker portion of the vesicle wall, its floor, being directed toward the mesometrial border. During the early hours of the seventh day, cell proliferation, cell rearrangement, and enlargement of cells takes place in the region of the germinal disc, resulting in a marked thickening of this portion of the wall of the vesicle, manifested by an outward growth, as also a growth inward into the cavity of the vesicle, initiating the phenomena known as 'inversion of the germ layers,' or 'entypy of the germ layers.' In this thickening of the germ disc, there may be recognized on the one hand the anlage of the ectoplacental cone or 'Träger,' on the other hand, in the cell mass which extends into the cavity of the blastodermic vesicle, the anlage of the egg-plug or egg-cylinder. In the anlage of the egg-cylinder there may be recognized early a circumscribed compact mass of cells, staining somewhat more deeply, which mass of cells I have designated the ectodermal node, since it is the anlage of the primary embryonic ectoderm of the future embryo. This ectodermal node, so far as it extends into the blastocele, is covered by the single layer of yolk entoderm, or as it is now known, the visceral layer of the entoderm. The ectodermal node is readily differentiated from the cells of the ectoplacental cone, with the base of which it is in close relation.

The more complete development and differentiation of the egg-cylinder, the anlage of which was noted during the seventh day, may be observed during the eighth day after insemination. The thin walled portion of the vesicle, its roof or antimesometrial portion, enlarges, assuming a distinctly cylindric form. The ectodermal node with covering of the layer of visceral entoderm is forced into the cavity of the vesicle, this by reason of proliferation of the cells at the base of the ectoplacental cone, this resulting in the formation of a nearly

cylindrically formed column of compactly arranged, polyhedral cells, interposed between the ectodermal node and the base of the ectoplacental cone, but merging into the latter without sharp, demarkation. To this column of cells, the name of extraembryonic ectoderm is given. The ectodermal node and the extraembryonic ectoderm together form a cylindric structure, surrounded by a single layer of visceral entoderm, which reaches from the base of the ectoplacental cone to nearly the mesometrial end of the original segmentation cavity. During the latter half of the eighth day, a small cavity appears in the ectodermal node. This is the anlage of the mesometrial portion of the proamniotic cavity. The cells bounding this cavity, derived from the cells of the ectodermal node, constitute the primary embryonic ectoderm. Soon after the anlage of the mesometrial portion of the proamniotic cavity, several discrete spaces become evident in the extraembryonic ectoderm of the egg-cylinder, constituting the anlage of the antimesometrial portion of the proamniotic cavity, these discrete spaces quickly joining to form a single space, the antimesometrial portion of the proamniotic cavity, lined by a single layer of cells of the extraembryonic ectoderm. Toward the end of the eighth day the mesometrial portion of the proamniotic cavity, arising in the ectodermal node, and the antimesometrial portion of the proamniotic cavity, arising in the entraembryonic ectoderm, fuse to form a single proamniotic cavity, the mesometrial portion of which is lined by primary embryonic ectoderm, the antimesometrial portion of which is lined by extraembryonic ectoderm, the two types of ectoderm forming a continuous layer, their line of union, however, being readily distinguishable. This hollow ectodermal cylinder, attached to the base of the ectoplacental cone and extending to the antimesometrial end of the segmentation cavity, is surrounded by a single layer of visceral entoderm in the meantime differentiated into a portion which surrounds the primary embryonic ectoderm, which consists of flattened cells and is now known as the primary embryonic entoderm, and a portion surrounding the extraembryonic portion of the egg-cylinder, consisting of tall columnar cells with vacualated protoplasm containing hemoglobin granules, and constituting an embryotrophic layer.

This cylindrical structure presents during the early part of the ninth day after insemination no evident bilateral symmetry, so that longitudinal sections, cut in planes at right angles to each other, present identical pictures. This is also evident in cross sections of the vesicles.

During the middle and latter part of the ninth day, active cell proliferation in the embryonic ectoderm in the region of the future caudal end of the embryo, leads to a distinct thickening of the embryonic ectoderm of this region. This thickening constitutes the primitive streak region. In it, there is developed a short axial groove, the primitive groove. From the edges of this groove, cells derived from the embryonic ectoderm, wander between ectoderm and embryonic entoderm. This constitutes the anlage of the mesoderm. There is no evidence of the participation of the embryonic entoderm in the anlage of the

mesoderm. Toward the end of the ninth day, and beginning of the tenth day after insemination, as a result of proliferation of the cells of the mesodermal anlage and further outwandering of cells from the embryonic ectoderm in the region of the primitive groove, the mesoderm extends so as to form a distinct layer situated between the two primary germ layers.

26. The development of the lymphatic drainage of the anterior limb in embryos of the cat. Lantern. GEORGE S. HUNTINGTON, Columbia University.

Two phases of the functional adaptation of early mammalian lymphatics are considered:

1. Since Miller's discovery in 1913 (Am. Jour. Anat., vol. 15, pp. 131–198) of the haemophoric function of the avian thoracic ducts in the early stages of their development, attention has been directed toward the determination of homologous conditions during lymphatic ontogeny in embryos of the other amniote classes. In the mammal (cat) the area of the jugular lymphsac and of some of its tributaries offers in the early stages conditions corresponding to those of the bird, although they are more obscure by reason of the close association with the adjacent systemic veins, a difficulty not encountered in the avian axial lymphatic line. The interpretation of mammalian lymphatic ontogeny gained from the viewpoint of the functional adaptation of early lymphatic channels serves to clarify some heretofore doubtful points in the mutual relations of developing lymphatic and venous channels in the mammalian embryo.

The vessel which offers the least complicated and clearest view of the genetic processes involved is the primitive ulnar lymphatic, draining the lateral body wall and the anterior limb bud, and accompanying the primitive ulnar vein during the period of the latter's functional activity, prior to the establishment of the definite subclavian venous line.

The first anlages of the developing primitive ulnar lymphatic are found in embryos between 7 mm. and 8 mm. as a disconnected series of intercellular mesenchymal spaces which form dorsal to the primitive ulnar vein and to the lower cervical nerve trunks. At first these spaces are irregular and communicate with smaller intercellular clefts in the surrounding mesenchyme. Later, in embryos of 8 mm. to 8.5 mm., they become distended and in part bounded by flattened mesenchyme cells. In embryos of 8.5 mm. to 9 mm. the originally separate individual spaces have united to form a continuous channel whose cephalic extremity effects a secondary junction with the dorsal division of the jugular lymphsac. This stage is usually completed in the 9 mm. embryo in which the primitive ulnar vein is paralleled at a little distance along its dorsal or dorso-lateral aspect by the distinct channel of the primitive ulnar lymphatic. The mesenchyme surrounding this vessel exhibits at numerous points groups of developing bloodcells. In the 9.5 mm. embryo this local haemopoesis attains its fullest develop-

ment and the red bloodcells begin to crowd the previously clear lumen of the primitive ulnar lymphatic channel. The walls of the latter appear to be formed still in part by undifferentiated mesenchymal cells, in part by flattened cells assuming distinct endothelial characters. At numerous points the lumen of the lymphatic channel is still in open communication with smaller intercellular clefts in the surrounding haemopoetic mesenchyme. Some of these enlarge to include groups of bloodcells and become added to the main lymph channel. The majority of red cells appear to gain access to the latter through these avenues. It is of course possible that some of the blood-contents of the ulnar lymphatic are due to reflux from the jugular lymphsac, but the conditions in the 9.5 mm. embryo seem to point clearly to the inclusion of the cells in situ in the manner described.

In the 10 mm. and 11 mm. embryos the primitive ulnar lymphatic appears enlarged, lined by a definite and closed endothelium and the lumen densely crowded with red blood-cells. This is the fully developed haemophoric stage of the vessel.

In embryos of 11.5 mm. evacuation of the blood contents of the primitive ulnar lymphatic into the jugular sac and through the same into the precardinal vein occurs. This process is usually completed in the 12 mm. and 12.5 mm. stages. The proximal segment of the primitive ulnar lymphatic rapidly narrows after evacuation is completed and in embryos of 13 mm. to 14 mm. the continuity of the channel becomes interrupted a short distance caudad to its point of entrance into the jugular sac. The endothelial cells lining the lumen become larger, more rounded, stain deeply and appear to revert to the indifferent mesenchymal type, obliterating finally all trace of the former channel at this point. This may occur as early as the 13 mm. stage or be deferred to the 14 mm. or even the 15 mm. stage.

2. After the interruption of the primitive ulnar lymphatic at the level stated above, the distal segment of the channel enlarges rapidly by distension with clear fluid and by the addition of numerous new intercellular spaces forming in the surrounding mesenchyme. In this way a vast lymphatic reservoir, the axillary lymphsac, is formed which for a time receives and stores the lymph drained from the limb-bud and body wall. The former path of lymphatic drainage of this area via the primitive ulnar lymphatic *dorsal* to the nerve trunks of the anterior limb into the jugular lymphsac having been interrupted, as above described, a new *ventral* lymphatic line is now established by the concrescence of numerous originally separate lymph spaces developed along the course of the recently established subclavian vein. The resulting channel connects cephalad with the ventral process extending caudad from the subclavian approach of the jugular lymphsac over the ventral face of the jugulo-subclavian venous angle. Distally it opens into and drains the axillary sac which subsequently becomes reduced in extent and incorporated in the permanent thoraco-appendicular lymphatic system. The axillary sac thus functions as a temporary storage reservoir for the lymph pending the completion of the change from

the *dorsal* primitive ulnar to the *ventral* permanent subclavian line of lymphatic drainage of the anterior limb and lateral body wall.

The definition of stages given above is based on observations covering a large number of closely graded embryos of the cat and represent the average. Considerable individual chronological variation is encountered. The material used comprises the following series of the Columbia University Embryological Collection in transverse section:

Cat 7.0 mm.	Serial No. 105, 108, 119, 121, 135, 137, 138, 266, 281, 487, 488, 752
Cat 7.5 mm.	Serial No. 282, 284, 486, 567, 595
Cat 8.0 mm.	Serial No. 89, 102, 485, 596, 597, 703
Cat 8.5 mm.	Serial No. 285, 466, 490, 704
Cat 9.0 mm.	Serial No. 106, 136, 265, 268, 421, 458, 459, 462, 467, 468, 489, 491, 492, 589
Cat 9.5 mm.	Serial No. 132, 133, 239, 269, 273, 461, 497, 499, 501, 598, 599
Cat 10.0 mm.	Serial No. 79, 101, 111, 112, 113, 114, 140, 237, 272, 274, 474, 477, 478, 496, 498, 500, 707
Cat 10.5 mm.	Serial No. 81, 118, 120, 479, 480, 720
Cat 11.0 mm.	Serial No. 77, 98, 213, 473, 566, 718, 719
Cat 11.5 mm.	Serial No. 251, 256, 472
Cat 12.0 mm.	Serial No. 78, 97, 100, 217, 263, 471, 744
Cat 12.5 mm.	Serial No. 264, 590, 591, 592
Cat 13.0 mm.	Serial No. 92, 107, 262
Cat 13.5 mm.	Serial No. 76, 189, 223
Cat 14.0 mm.	Serial No. 122, 127, 210, 211, 212, 214, 747

The paper is illustrated by photomicrographs of the sections and Lumière lantern slides of the reconstructions.

27. Effect of acute and chronic inanition upon the relative weights of the various organs and systems of adult albino rats. C. M. Jackson, Institute of Anatomy, University of Minnesota, Minneapolis.

Twenty-one well-nourished adult rats were used, initial body weights varying from 182 to 367 grams. Fifteen rats were used for acute inanition, being allowed water but no food. They were killed after 6 to 12 days, the loss in body weight varying from 25 to 39 per cent (average loss, 36 per cent). Six rats were subjected to chronic inanition, being underfed so as to reduce the body weight slowly through a period of about five weeks, and were killed when the loss in body weight reached about 36 per cent. The results below stated apply in general to both acute and chronic inanition, unless otherwise specified. The published data of Donaldson, Hatai, Jackson and Lowrey are taken as normal for comparison. On account of the great variability of some organs and the relatively small number of observations, final conclusions are in some cases uncertain.

The head and fore-limbs lose relatively less than the body as a whole. Their relative (percentage) weight therefore increases. Of the systems —integument, skeleton, musculature, viscera and 'remainder'—the in-

tegument loses relatively nearly the same as the whole body, and therefore nearly maintains its original relative (percentage) weight. The same is true of the musculature, which however undergoes a somewhat greater loss in relative weight during chronic inanition. The visceral group, as a whole, undergoes little change in relative weight, decreasing slightly in acute inanition. The individual organs, however, vary greatly, as indicated below. The skeleton retains nearly its original absolute weight, and therefore increases greatly in relative weight. There is a corresponding decrease in the 'remainder,' due chiefly to loss of fat.

The individual viscera may be classified in three groups:

(1) The brain, spinal cord and eyeballs show little or no loss in absolute weight, compared with the normal at the initial body weight, hence their relative (percentage) weight has markedly increased with the diminution in body weight during inanition. The same is apparently true for the thyroid gland in acute inanition, but in chronic inanition there is apparently a loss, though relatively less than in the body as a whole. The suprarenal glands also apparently lose less in absolute weight than the body as a whole, hence their relative (percentage) weight increases.

(2) The heart, lungs, kidneys, testis, epididymis and hypophysis undergo nearly the same relative loss in weight as the body as a whole, therefore their relative (percentage) weight remains nearly the same. The thymus has already undergone age involution, and is therefore not materially affected by inanition.

(3) The liver and the alimentary canal (both empty and including contents) usually decrease in weight relatively more than the body as a whole. The spleen is exceedingly variable; in acute inanition it usually shows a marked decrease in relative weight (although averaging higher in chronic inanition).

28. Changes in young albino rats held at constant body weight by underfeeding for various periods. C. M. Jackson, Institute of Anatomy, University of Minnesota, Minneapolis.

Ten litters, including 65 rats, were used. Twenty-five rats were used as controls, including 11 at 3 weeks of age, 2 at 6 weeks, 6 at 10 weeks, 3 at 32 weeks, and 3 at 35 weeks. In addition, data previously gathered by observations upon several hundred normal rats were available for comparison. Forty rats were held at constant body weight by underfeeding for various periods, 8 rats from age of 3 weeks to age of 6 weeks; 3 rats from 3 weeks to 8 weeks; 22 rats from 3 weeks to 10 weeks; 1 rat from 3 weeks to 13 weeks; 1 rat from 3 weeks to 16 weeks; 2 rats from 6 weeks to 32 weeks; and 3 rats from 10 weeks to 35 weeks. On account of the great variability of some organs, the number of observations in some cases is insufficient for final conclusions.

As to body proportions, the relative weights of the head, trunk and extremities remain practically unchanged in young albino rats held at constant body weights.

Of the systems—integument, skeleton, musculature, viscera and

'remainder'—there is but little change in the weight of the musculature, visceral group (as a whole) and 'remainder.' There is, however, usually a marked decrease in the integument, counter-balanced by a marked increase in the skeleton. Thus under these conditions the growth capacity appears weakest in the skin and strongest in the skeletal system. This is in striking contrast with the normal growth process, during which the musculature shows the greatest increase and the skeleton lags behind relatively.

The increase in the skeleton during constant body weight appears to involve the ligaments and cartilages as well as the bony skeleton. The skeletal growth tends to proceed along the lines of normal development, as indicated by changes in water content, by formation and union of epiphyses, and by relative elongation of the tail (compared with trunk length). The teeth also continue to develop normally (eruption of third mars).

The individual viscera may be classified in three groups.

(1) There is during the maintenance of constant body weight a well-marked increase in the weights of the spinal cord and eyeballs; usually also of the tetis alimentary canal (both empty and including contents) and hypophysis.

(2) There is no marked change in the weights of the brain, heart, lungs, suprarenal glands, kidneys and epididymi. The liver is variable, with apparently a slight tendency to increase in the younger rats and to decrease in the older.

(3) There is always a marked decrease in the weight of the thymus ('hunger involution'); usually also of the spleen (earlier stages) and probably to a slight extent of the lungs and thyroid gland.

29. Haemopoiesis in the yolk-sac of the pig embryo. H. E. Jordan, University of Virginia, Va.

The yolk-sac of the 10 mm. pig embryo was found to be especially favorable for a study of the early stages in blood-cell formation. It is still sufficiently young to show the earliest steps (with the exception of the initial origin of the angioblast) and sufficiently advanced to include the more important of the later stages. For these reasons it seems desirable to limit the present description to this stage of development. It need simply be added that the haemopoietic phenomena are essentially the same in the yolk-sacs of specimens ranging from 4 to 12 mm. Still younger and older specimens have not yet been examined.

This description pertains chiefly to a specimen fixed in Zenker's fluid and stained in toto with Delafield's hematoxylin and counterstained with eosin. The specimen is exceptional only in its unusually good preservation. Mitotic figures are abundantly present in all of the tissues of the embryo and the sac; and the mitochondrial content of cells of the liver and the entoderm of the sac are clearly shown. It would seem that the specimen may therefore be confidently regarded as perfectly normal and well preserved.

The point of special importance pertains to the evidence for the origin of primitive blood cells or haemoblasts, from the mesenchyma. This

is the link in the monophyletic view of haemopoiesis as urged notably by Saxer, Bryce, Maximow and Dantschakoff concerning which there remains perhaps the greatest doubt. In the belief that the yolk-sac offered the best material for an investigation of this particular point, this study was undertaken. Both direct and indirect evidence strongly indicates that the mesenchyma of the young yolk-sac does differentiate into blood-vascular tissue, or so-called angioblast.

Regarding the origin of the initial angioblast in the yolk-sac of the pig this material yields no data. To identify angioblast with mesenchyma, at least in part, in the yolk-sac of this stage may seem to beg the entire question. Against this objection can be brought the observation that in certain portions the entire extra-entodermal layer forms a continuous tissue, or syncytium. The continuity is complete only in the location of early blood-islands; where blood vessels appear the continuity pertains to the endothelium. Moreover, there is apparently no difference, either from the standpoint of cytoplasmic or nuclear structure or form, between endothelial cells, mesenchymal cells, and the surface mesothelial cells. And in certain regions the three are seen to be continuous. The criteria which Clark (Anat. Rec., vol. 8, no. 2; 1914) used with success in the chick embryo for the differentiation of endothelial from mesenchymal cells are not applicable to this material. On the other hand, the angioblast is everywhere sharply delimited from the entoderm. The morphologic evidence seems to force the conclusion that endothelium (angioblast), mesenchyma, and mesothelium are at this stage composed of the same cell, slightly modified along different lines by chiefly mechanical factors. In mesothelium and endothelium these factors include predominantly the element of pressure forcing an elongation and flattening of the cells. This accounts for the close similarity, amounting apparently to an identity, between the endothelial and the mesothelial cell.

Since it can be readily proved, as will be shown below, that haemoblasts differentiate from the endothelium of these early blood-vessels, these observations regarding the similarity and continuity of endothelium and mesenchyma constitute the indirect evidence for the mesenchymal origin of primitive blood cells. That mesothelium and endothelium, in spite of their histologic close similarity, are however functionally different at this stage must be admitted from the fact that no evidence appears of a direct origin of haemoblasts from mesothelium. But the continuity of mesothelium and mesenchyma must again be emphasized, as well as the proliferative capacity of the mesothelium. Moreover, that mesothelium may function haemopoietically is shown in Bremer's description of mesothelial ingrowths (angioblast-cords and angiocysts) into the body-stalk of a young human embryo (Amer. Jour. Anat., vol. 16, no. 4, 1914).

The direct evidence for the origin of haemoblasts (primitive lymphocytes—Maximow; mesamoeboid cells—Minot) from mesenchyma appears chiefly in the presence of certain cells in the mesenchymal syncytium with the cytoplasmic and nuclear features of true intra-vascular haemo-

blasts and megaloblasts. There can be no doubt regarding their identity. What needs to be established is their true relationship to the mesenchymal cells. It is possible that they may have wandered into the mesenchyma from the blood vessels. This is Minot's suggestion regarding the 'lymphocytes' which Maximow has described as arising in the body-mesenchyme of the young rabbit embryo; and he bases his conclusion upon the observation that such cells frequently show certain nuclear and cytoplasmic features which he interprets as degenerative (Keibel and Mall's Human Embryology, p. 511, vol. 3). The cells in question in the yolk-sac of the pig show no nuclear or essential cytoplasmic differences which seem to warrant the interpretation that they are degenerating cells.

But neither these facts nor the additional one, namely, that they appear to be continuous through numerous processes with the mesenchyma, prove that they have arisen *in situ*. The processes may be of the nature of those described by Kite (Journ. Infect. Dis., vol. 15, no. 2, 1914) for polymorpho-nuclear leucocytes under certain conditions; and these pseudopodia of a possible wandering cell may have become so intimately fused with the mesenchymal syncytium as to simulate continuity. The following observation, however, seems to establish the *in situ* origin of these haemoblasts in the mesenchyma: In the case of certain undoubted haemoblasts which are still in continuity with an undoubted mesenchymal cell, a delicate chromatic thread attached to the haemoblast nucleus extends for a considerable distance through the connecting bridge of protoplasm towards the nucleus of the mesenchymal cell. In a few instances the connection was apparently complete. This chromatic bridge is always most conspicuous at its haemoblast terminal, and looks like an evagination from the haemoblast nucleus. That this chromatic bridge indicates amitotic division is doubtful, since many of the mesenchymal cells are seen in mitosis. But whatever its interpretation in terms of cell division, it would seem to show a haemoblast origin from mescenchyme. In some instances the evidence indicates that the mesenchyme becomes arranged in the form of endothelium about such forming haemoblasts. Haemoblasts are occasionally seen in process of transit between mesenchyma and blood vessel, the direction being probably either way. Certain spaces in the mesenchyme are lined by flattened endotheliod (mesothelial) cells.

It remains to describe blood cell origin and differentiation within the blood vessels. The blood vessels are at the start simply endothelial tubes of irregular caliber. The blood cells within the vessel include haemoblasts, cyrthroblasts (megaloblasts and normoblasts) and giant cells. No evidence appears of leucocytes except in so far as the smaller basophilic haemoblasts simulate lymphocytes. All of these cells are capable of intense proliferative activity.

The *haemoblast* (mesomoeboid stage of Minot) is a relatively small spherical cell with a relatively large nucleus, and a narrow shell of slightly basophilic cytoplasm. The nucleus contains a delicate wide-meshed chromatic reticulum with generally several larger spheroidal

chromatic masses. Occasional cells answering to this description are of much greater size.

The *megaloblast* (ichthyoid stage of Minot) phase of the developing erythroblast is a relatively much larger cell. Its nucleus, however, is approximately of the same size and structure as that of the haemoblast. Its considerable cytoplasmic body reacts to the eosin stain. It thus has a bright pink color, due presumably to the presence of a small amount of haemoglobin. These cells divide extensively; they present a large series of size variations; moreover, the larger parent cells are frequently of lenticular or bluntly fusiform shape. This peculiar shape is interpreted in terms of an endothelial origin, as will be described below. Again, certain cells of this type possess a number of blunt pseudopodia. Certain cells with bilobed nuclei suggest that the nuclei of certain of these cells may also occasionally divide by amitosis.

The mormoblast (saunoid stage of Minot) is also a relatively large cell, with a relatively smaller, denser, more chromatic granular nucleus. This is the most abundant type of cell. It is very uniform from the view-point both of size and structure. Very many of these cells are seen in mitosis. The cytoplasm of this cell is peculiar; apparently the technic employed extracted the haemoglobin; the cytoplasm appears clear, with a very wide-meshed delicate reticulum. A smaller number of cells are present very similar to the normoblasts, except that the nucleus is still smaller and more chromatic. This is the *erythrocyte* in early stage of metamorphosis into an *erythroplastid*. This involves the extrusion of the nucleus together with an enveloping shell of cytoplasm, as described by Emmel (Am. Jour. Anat., vol. 16, no. 2, 1914). This stage is extremely rare, but in view of Emmel's careful work can be properly interpreted.

The *giant cells* are relatively enormous cells, and very variable from the standpoint of the number, size and chromaticity of the nuclei. The larger varieties have a more deeply staining apparently cytoplasm. A graded series can be traced from the megaloblast with one nucleus, through one with two nuclei still retaining a pink-staining cytoplasm, to cells with more nuclei (or a larger, more chromatic nucleus) and a deep-brown-staining cytoplasm. This indicates the origin of the giant cells, namely, through growth (frequently accompanied by endogenous multiplication of the nucleus) of a megaloblast (or haemoblast).

From the standpoint of nuclear material giant cells have relatively little cytoplasm. From this viewpoint they represent young or 'rejuvenated' cells. There is some evidence to indicate amitotic division of the nuclei; but mitotic figures also appear, frequently with tri-and multipolar spindles. All the evidence indicates very rapid growth of these cells. They are of two types, mono- and multinucleate. As to their function the evidence seems clear in the case of the multinucleate type. The megakary-ocytes most probably subsequently become multinucleate through division of the nucleus, and thus partake of the same function. About one or several of the nuclei appear clearer courts

identical in structure with that described for the normoblast. The nuclei meanwhile also assume the features of the normoblast type. Where in a binucleate cell one of the nuclei and its surrounding cytoplasm is thus modified, while the remainder of the cell retains the features of the megaloblast, the appearance might be interpreted in terms of the ingestion of a normoblast by a megaloblast. But where both nuclei of the same cell, as is frequently the case, and their adjacent cytoplasmic areas are similarly differentiated, the interpretation is in applicable. The polykaryocytes of the yolk-sac undoubtedly have at least as a partial functional rôle that of the formation of normoblasts as here described.

In the human yolk-sac Spee (Anat. Anz., Bd. 14, 1896) described the giant cells (to which he also attributed a haemogenic function) as arising from the entodermal cells. This view is untenable for the yolk-sac of the pig. Giant cells and the entodermal cells lining the sac have no features in common except their general staining reaction. The cytoplasm of the entodermal cells contains distinct basal filaments (mitochondria). Such are absent in the giant cells. Also, the entodermal cells contain a single relatively small nucleus, with coarse chromatic net, and usually two large spherical chromatic nucleoli. Moreover, there is never an intimate spatial relationship between entodermal and giant cells, and nothing appears in the nature of transition stages.

In passing may be noted the very close similarity between the entodermal cells of the yolk-sac and those of the liver. Graf v. Spee ('96) and also Paladino ('03) have attributed to the entoderm of the yolk-sac an hepatic function on the basis of a general morphologic similarity. This similarity in the pig pertains even to the finer cytoplasmic structure and content, namely apparently identical mitochondrial threads. It should be noted that no other tissues in this specimen showed distinctly any mitochondrial elements. This statement is not meant to imply that they were not present—there is sufficient recorded evidence to prove that they are—but only that this technic did not preserve them, while it reveals very beautifully the special threads in the entodermal cells of the sac and the liver cells.

Similar deep-staining filament were described by myself (Anat. Anz., Bd. 31, 00, 11, 1907, and Bd. 39, 00, 1, 1910) in the yolk-sac of a 9.2 mm. human embryo as 'mucinous masses,' and subsequently by Branca (Ann. de Gynec. et d'Obst., tome 2, 1908) in yolk-sac of about the same age as 'functional protoplasm' (ergastoplasm). They are very similar to the ergastoplasmic filaments of certain secretory cells as, for example, those of the pancreas, where they have been described as segmenting distally into "secretory granules." No such segmentation is discernible in these cells from the yolk-sac of the pig. In the human yolk-sac the entodermal cell contained a generous irregular granular content but the granules showed no direct relationship to the filaments, and they were tentatively interpreted as débris incidental to degenerative changes. Mislawsky (Arch. Micr. Anat., Bd. 81, 00, 4, 1913) has recently shown by aid of delicate differential staining methods that the

basal filaments of the pancreas cell are in reality condrioconts, and have nothing directly to do with formation of secretion granules. These filaments of the yolk entoderm and hepatic cells are more probably of the nature of mitochondria and give evidence of the metabolic virility of these cells.

It is of importance also to note that all the types of blood cells described for the yolk-sac are present also in the liver, only the normoblasts are relatively much more abundant. Elsewhere in the embryo (including especially the heart) normoblasts and later erythrocytes are exclusively to be seen.

Finally, as to the rôle of the endothelium of the yolk-sac vessels in haemopoiesis: the matter may be summed up by simply stating that endothelial cells differentiate into haemoblasts, both intravascularly and to a slight extent also extravascularly. This involves a rounding up of the cytoplasm about an endothelial cell nucleus and a subsequent abstraction of the forming cell from the wall of the vessel. Such process is probably commonly preceded by proliferation of the endothelial cell involved. Mitosis of endothelial cells are fairly abundant. Occasional endothelial cells are binucleated. The haemoblast need not necessarily at once separate from the endothelial wall. It may retain its connection through the succeeding stages of development including the fully differentiated megaloblast; and there is some evidence to show that even a giant cell may thus remain connected. The evidence for this, not however quite complete in the case of the giant cell, consists in a graded series of developmental stages from the true endothelial cell to the megaloblast. The megaloblast of this series is of lenticular form, its pointed terminals passing into a thread of protoplasm continuous in both directions with the endothelial wall.

30. Morphological evidences of intracellular destruction of red blood-corpuscles. Preston Kyes, from the University of Chicago.

The intracellular destruction of red blood-corpuscles by vascular endothelium although recognized as of frequent occurrence under pathological conditions, has not been established as a physiological process taking place under normal conditions.

It is the purpose of this communication to give morphological evidences that in the case of the pigeon, among other species, there is a constant normal phagocytosis of red blood-cells by specialized vascular endothelium in the liver and spleen.

Sections of suitably fixed tissue from the liver and spleen of pigeons display when subjected to Perl's test for iron, a distinctly differentiated type of cell marked by the Prussian blue tone of the cytoplasm due to a positive iron reaction. Combined with counter-stains, therefore, Perl's method affords a favorable technique for microscopic study of the cells in question, which cells will be interpreted later as phagocytic endothelial cells whose iron content is due to ingested red blood-corpuscles. In detail, the histological technique which I have employed in studying the tissues of eighteen normal pigeons, is as follows:

Fix thin slices of tissue for 18 to 24 hours in Muller's fluid plus 5 per cent mercuric sublimate. Imbed in paraffin and section to 4 microns. Fix sections to slide and stain 20 to 40 minutes with acid carmine. Wash, and transfer to equal parts of a 2 per cent aqueous solution of potassium ferrocyanide and of a 2 per cent aqueous solution of hydrochloric acid. Remove after from 3 to 10 minutes, wash in distilled water and pass quickly through a 0.5 per cent aqueous erythrosin solution. Dehydrate in alcohol, clear in xylol and mount in Canada balsam.

Specimens of normal pigeon's liver and spleen prepared according to the above method, display an extensive content of cells possessing the distinct blue tone of the Prussian-blue iron reaction. These cells are distributed rather evenly throughout both organs but more numerously in the liver. Under low powers of the microscope their general morphology indicates that the iron-containing cells are of the same type in the two organs, and as will be seen later this is supported by a correspondence in their finer structure and physiology. Inasmuch as the relation of these cells to other structures is much more evident, however, in the liver, their first description is limited to that organ.

In liver specimens observed under medium magnification, the cells referred to above appear as blue patches sharply differentiated from the red-stained parenchyma. These cells are larger in their greatest dimension than the liver cells proper, vary much in size and form, and are often seen to contain two or three carmine—or eosin-stained bodies. In their distribution they display a constant relation to the venous capillaries, often appearing to occupy the lumen of these vessels. Under the higher powers of the microscope, it is seen that each cell is an integral part of the endothelial intima lining the capillaries; in other words a fixed tissue cell engaged by one of its surfaces upon the reticulum of the vessel wall and with a free surface bulging to a greater or less degree into the vessel lumen. The attached surface of the cell follows strictly the line of the vessel-wall be it straight or curved, often continuing around an angle of bifurcation. No processes are seen extending between the liver cells: in fact I have not seen evidence that these cells possess processes extending in any direction. The cells under discussion are clearly those described in the liver of mammals by v. Kupffer first as perivascular connective-tissue cells and finally as intimal cells. To these cells the terms 'Sternzellen,' 'stellate cells,' 'Kupffer cells,' have been applied in reference to the liver; but to include the same cell as also seen in the spleen and where not, I employ the term hemophage.

The nucleus displayed by the hemophage stains a deep garnet with the carmine used in the given technique and contains two or three very distinct and intensely stained nucleoli. In the hemophages which are more nearly flat, the nucleus appears as those of the typical endothelial cell, whereas in the protruding cells of greater bulk, the nucleus is more vesicular and is irregularly pyramidal in form. Two nuclei may be found within a single cell. This but rarely, however.

The most striking characteristic of the hemophage is the morphology of its cell-body. This is determined by the fact that within vascuoles

of the cytoplasm, are contained red blood-corpuscles taken from the circulating blood stream. It is not meant that here and there may occasionally be found a hemophage which has taken up an erythrocyte, but rather, that the occurrence is general and that approximately one-third of the total intimal cells are active hemophages and that each hemophage displays evidence of containing, or having recently contained, one or more erythrocytes. The fact that the red blood-corpuscles of birds are nucleated, have a definite ovoid outline, and are of relatively large size, allows clear observation as to their actual inclusion and ultimate intracellular fate. The cell-body of the hemophage has no fixed morphology but changes from time to time according to the phase of its phagocytic activity. Within a single field of the microscope may be seen all intermediate stages between the hemophage whose cell-body is greatly distended by an intact erythrocyte recently ingested and the hemophage which has so far completed the destruction of the erythrocyte as to again appear as a flat endothelial cell except for the presence of the traces of the end-products of the digestion. In the first instance the cell-body of the hemophage bulges markedly into the capillary lumen and its nucleus is crowded to one side. The included erythrocyte in this earliest stage appears in all ways the same as those of the blood-stream and displays the normal staining reaction; namely, by the technique given, its nucleus stains a deep red-brown, while its cytoplasm stains an even yellow bronze tone. In hemophages which represent the subsequent stages, the included erythrocytes are seen in various stages of disintegration and digestion while the cytoplasm of the including cell gives a constant iron reaction. The first marked change in the erythrocyte is hemolysis, the hemoglobin escaping into vacuoles of the cytoplasm of the phagocytic cell and leaving the nucleus-containing stroma distinctly outlined. Gradually both the stroma and nucleus lose their staining reaction, until finally the vacuole contracts about a small indistinct remnant of the nucleus which in its turn ultimately disappears.

Meanwhile the hemoglobin which has escaped into the cytoplasm of the hemophage is seen to undergo a series of changes. At first the greater part of the pigment does not give the iron reaction but retains its yellow-bronze tone with erythrosin and occupies vacuoles of various sizes. In hemophages representing a later stage, however the contents of the vacuoles also give the iron reaction and with great intensity, contrasting with the lighter blue of the surrounding cytoplasm. Such cells finally show no content of unmodified hemoglobin. With the disappearance of the native hemoglobin, therefore, there is a parallel increase in the iron-reacting pigment. In untreated specimens this pigment is golden-yellow and is presumably hemosiderin. The hemophages which represent the last stages in the phagocytosis and digestion, appear less and less bulky, with a fainter iron reaction and a less vesicular nucleus. The last observable stage is represented by a cell which contains no yellow pigment but which in all ways appears as a typical endothelial cell of the vascular intima except, however, that its cytoplasm gives a faint and diffuse iron reaction.

As stated above, examples of the stages just outlined are readily seen in a single microscopic field and the interpretation of the sequence of events which they represent leads to the conclusion that the cells of the vascular endothelium of the venous capillaries of the liver of birds in performing a normal physiological function, ingest intact red-blood corpuscles, hemolyse the same, destroy the stroma and nucleus, split the hemoglobin with a freeing of the iron, and finally return to their original form.

In the spleen, the hemophages are seen in distinctly fewer numbers than in the liver. For the most part they are confined to the pulp cords in contrast to the Malpighian follicles and have no such evident relation to a vessel-wall or lumen as in the liver. The hemophage, however, is morphologically in all of its details of the same type as that of the liver, and the phases of ingestion and digestion of erythrocytes form the same cycle giving the same iron reaction at corresponding points.

With the recognition of a constant normal phagocytosis of erythrocytes by the intimal cells of the venous capillaries of the liver and corresponding cells in the spleen, the question arises as to how far these cells differ from vascular endothelium in general; in other words, the extent of their specialization. In reference to this point, the evidence shows that the phagocytosis is normally accomplished by endothelium in certain locations only. Thus in the liver, the hemophages are confined to the intima of the venous capillaries, while the intima of the larger vessels displays no such phagocytic action.

In applying the same technique in a study of the livers of the frog (Rana pipiens), toad (Bufo lentiginosus), turtle (Chrysemys marginata), crocodile (Alligator mississipiens s), and opossum (Didelphys virginiana), I have found a similar cycle of intracellular blood destruction in the corresponding cells of the reptiles, amphibia and mammals.

It would appear, therefore, that the application of a trustworthy differential histological method, shows that the liver and spleen of many species, do contain specialized endothelial cells which have as a normal physiological function the destruction of red blood-corpuscles with a liberation of the contained iron.

31. On the implantation and placentation in the Sciuroid rodents (*lantern*).

Thomas G. Lee, Institute of Anatomy, University of Minnesota.

In 1902 and 1903 the writer published descriptions of the implantation of the ovum in Spermophilus. In this work attention was called to a method of implantation and a series of structural changes preceding the formation of the true placenta which were unlike those of any other previously described mammal, and at the same time were the first account of the implantation in any of the Sciuroidae. These observations were confirmed on the European Spermophilus by Rejsek. In 1905 Müller found similar conditions in the European red squirrel, Sciurus. In 1910 the writer described the early stages of Cynomys at the International Anatomical Congress in Brussels. Since 1902 the writer has been engaged in collecting early stages of various genera of

American Sciuroid rodents to determine if the peculiar conditions found in Spermophilus (or 'Citellus' as the taxonomists have since decided upon as the proper generic name) were characteristic of this large group of rodents. The collection of very early stages of wild rodents which breed for the most part but once a year, and whose genera are widely separated over the United States, is an extremely tedious and very expensive undertaking, but sufficient material has been secured to date of the following genera of the Sciuroidae to determine that the general method of implantation and placentation as previously described for Citellus (Spermophilus) hold true for the larger division. The genera studied include Citellus, 3 species, Ammospermophilus, Tamias, Cynomys, and Sciurus. The writer is preparing a more complete description and illustration of the early developmental conditions characteristic of these Sciruoidae than was possible before with the quite limited material. The greater variety of material now available enables one to point out the interesting slight divergences among the several genera.

32. On the relationship of the endocardium to entoderm in Citellus. Thomas G. Lee, Institute of Anatomy, University of Minnesota.

In studying the early development of the sciuroid rodent Citellus the writer noted the following described conditions which may be of interest to investigators working on that yet unsolved problem of the origin of the vascular system.

With the folding over and fusion of the entodermal walls of the foregut to form the pharynx region, there is to be noted a dorso-lateral angle on either side formed by the dorsal wall on either side of the chorda and the lateral wall of the pharynx and a somewhat less prominent ventro-lateral angle or groove, which will be designated in this paper as the cardiac sulcus.

Each cardiac sulcus is a groove or furrow in the free surface of the entoderm which follows the course of the lateral hearts. It begins in the lateral wall of the mid gut region and extends forwards to enter the closed pharynx region at the ventro-lateral angle and then continues along the ventral wall of the pharnyx converging to unite with the opposite sulcus in the mid ventral line just above the point of fusion of the lateral hearts.

The entoderm in the line of the cardiac sulcus is considerably thicker than that on either side. This thickening, however, is not uniform; it is more pronounced in certain areas than others. There is thus produced a corresponding elevation or ridge of the entoderm in the direction of the lateral heart. This thickened entoderm constitutes the walls of the sulcus. The groove, while easily recognizable throughout its course, varies in its shape; in places it is narrow and deep, in other places it becomes widened out and quite shallow. In embryos of this stage of development, the fold of splanchnic mesoderm which will form the myocardium does not completely envelope the endothelial tube of the lateral heart, the interval being completed by the entoderm of the foregut. It is this portion of the entoderm that forms the

cardiac sulcus. The above described sulcus is not peculiar to Citellus but is figured by many investigators, as Kölliker in the rabbit, Fleischman in the cat, Bonnet in the dog. It is a transitory structure but is probably to be found in all mammals at the proper stage of development. While this region has been figured, almost no reference has been made to it as far as I am at present familiar with the literature.

In a number of series of Citellus I have found interesting examples of an intimate relationship between the entoderm of the cardiac sulcus and the endocardium of the lateral heart as shown by the reconstructions and drawings that illustrate this paper.

In the Citellus embryo here modelled, the primary implantation attachment (previously described by the writer) is just separating while the trophoblastic attachment for the allantoic placenta is beginning at the mesometrial portion of the uterine cavity; the amnion is not yet quite complete; the foregut is closed, the ectoderm of the oral plate is fused with the entoderm but not broken through; the pharynx does not yet show the evagination of the pouches; the endocardium of the lateral heart is beginning to fuse at the anterior end; the two dorsal aortae are well outlined, the first aortic arch is not yet completed. In an embryo at this stage the endocardium of the lateral heart on either side, in the region between the junctions of foregut and midgut and the point of beginning union of the two lateral hearts, shows an intimate relationship to the thickened entoderm of the cardiac sulcus. The endocardial tube is free and separate from the myocardial fold of splanchnic mesoderm in the sections, the contour of the tube is either oval or pear-shaped with a portion of the endothelial wall extended out as a thin fold or strand of cells toward the sulcus. Examining the series section by section it will be seen that while in each there is the extension of the fold or strand of cells toward the sulcus, in certain sections there is a short interval and in others very close contact; in certain sections there is distinct continuity with the entoderm of the sulcus walls. This intimate relationship is lost in the region of the fusion of the lateral heart.

33. The comparative embryology of the mammalian stomach. Frederic T. Lewis, Harvard Medical School.

The study here reported has been carried out in large part by Dr. C. H. Heuser of The Wistar Institute of Anatomy, and a preliminary report of it was presented by him at the last meeting of the Anatomists. The work has now been carried further, and is nearly finished. Four animals have been studied, the cat, rat, pig, and sheep. The simple lenticular stomach from which the very diverse adult forms proceed, has been modelled as a starting point (cat, 6.2 mm.; rat, 5.4. mm.; pig, 7.8 mm.; sheep, 7.2 mm.). Even at this stage there are some significant differences, notably in the decidedly convex lesser curvature in the sheep.

In the human stomach, the later development has been the subject of a paper already published by the writer. The effort is now made to obtain the stages of these other mammals most suitable for comparison. From a considerable number modelled, four stomachs of each species

have been chosen, ending with the rather definite stage in which the smooth epithelial lining has become corrugated.

In the cat, the simple carnivorous type of stomach is early manifest. There is a well-marked angular incisure separating the elongated cardiac portion from the tubular pars pylorica. The fundus is less prominent than in any of the other mammals chosen, including man, and the gastric canal, following the cardiac part of the lesser curvature, is relatively late in its development. As a whole the stomach is less differentiated than in any of the other forms. In the rat the fundus early becomes elongated, forming a capacious pouch with its tip hooked toward the oesophagus. The gastric canal is short, but well-defined, and ends at a distinct angular incisure. In the pig there is also a large fundus, which soon overhangs toward the right side. The gastric canal ends at an incisure which produces only a shallow indentation of the lesser curvature. Below it, as in the rat, the pars pylorica is at first capacious, apparently representing a pyloric vestibule, and then more tubular, forming the pyloric antrum. The sheep's stomach, at 10 mm., presents a slight angle indicating the lower end of the gastric canal, and a much deeper incisure below the rounded ventral swelling of the lesser curvature, which is the beginning of the future psalterium. The psalterium is an early and prominent subdivision in the sheep, but is scarcely indicated in the other forms. The fundus in the sheep develops into the large rumen. Even though it is lined with stratified epithelium in the adult, it cannot be regarded as a portion of the oesophagus, as some have taught. The reticulum represents the body of the stomach, and the abomasum is the pars pylorica, including both vestibule and antrum. In all of the forms examined the fundus, corpus and gastric canal are readily identified. The subdivisions of the pars pylorica require further study.

34. Reversed torsion of the human heart. Frederic T. Lewis, Harvard Medical School, and Maud E. Abbott, McGill University. Presented by Dr. Lewis.

While preparing the chapter on congenital cardiac disease for Osler's "Modern medicine," Dr. Maude E. Abbott, curator of the Medical Museum in Montreal, visited the Warren Museum and examined all the abnormal hearts which it contains. Among them is the heart of a man who died of phthisis in 1838, when twenty-one years of age. Its interventricular septum is so slightly developed that it was overlooked in the contemporary account of the specimen. The most interesting feature of this heart, however, is the large ventrally placed artery which appears to be the pulmonary artery, but which in reality is the aorta. It passes from the left side of the imperfectly divided ventricle toward the right, crossing the ventral surface of the pulmonary artery. The latter leaves the ventricular cavity like an aorta. Although these vessels have been cut away quite close to the heart, their distal relations as to aortic arch, right and left pulmonary branches and ductus arteriosus were apparently normal. Moreover, the atria and atrio-ventricular valves are normally arranged. In fig. 1B this heart is shown beside a normal one (fig. 1A) for comparison.

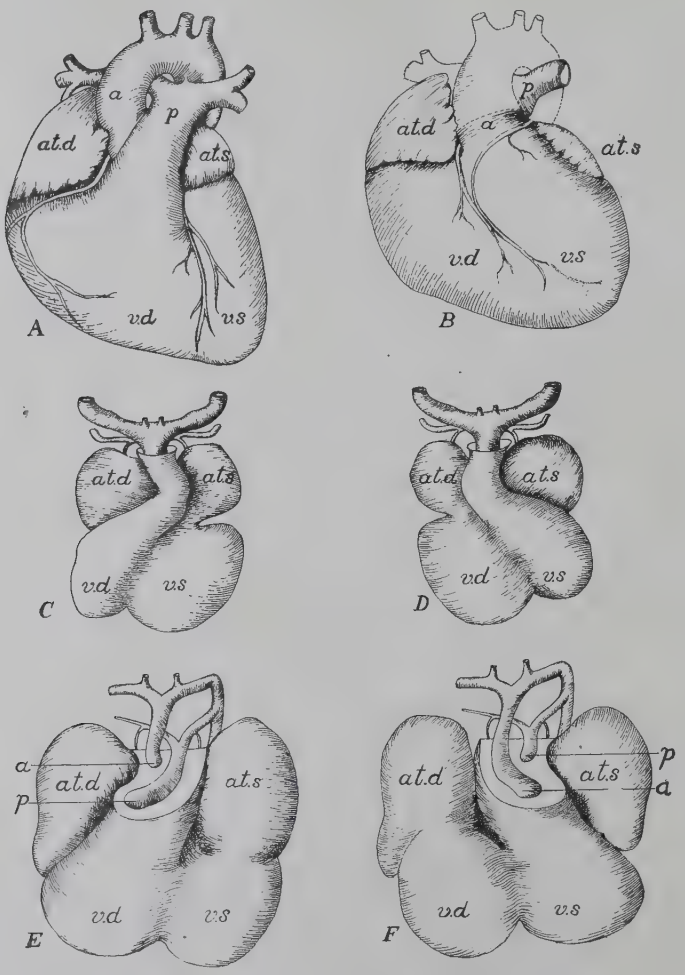


Fig. 1. *A*, ventral view of a normal adult human heart. *B*, corresponding view of an adult human heart showing reversed torsion. *C*, model of the heart of a 4.9 human embryo, which in *D* has been manipulated so as to present reversed torsion. *E*, model of the normal heart of a 10 mm. human embryo, the torsion of which has been reversed in *F*.

a, aorta. *at. d*, right atrium (or auricle). *at. s*, left atrium (or auricle). *p*, pulmonary artery. *v. d*, right ventricle. *v. s*, left ventricle.

Dr. Abbott brought this specimen to the writer for embryological interpretation, and he made the suggestion that in the embryo the cardiac tube had bent in the reverse direction to that which is normal, so that the aortic limb turned upward on the left side of the common ventricle instead of on the right. In order to verify this supposition, which we found had already been made by Keith, Dr. Abbott and the writer together undertook the following investigation. Normal embryonic hearts of the critical stages were selected and modelled as they occur in the embryo. Second models were then made, in which the ventricular portion of the heart was reversed, section by section. The two models of the heart of a 4.9 mm. embryo, kindly loaned to us by Dr. A. S. Begg, have been completed, and with others which are unfinished they seem to demonstrate the correctness of the interpretation. The distal part of the aortic trunk, including the roots of the pulmonary and fourth aortic arches, remains undisturbed in all its relations, and the atria and atrio-ventricular orifices are also in essentially normal position. But the reversal of the primary torsion causes the aorta to be split off from the truncus arteriosus ventral to the pulmonary artery, around the front of which it swings to the left ventricle. By manipulating the embryonic hearts in this way, the conditions in the abnormal adult specimen can be produced very satisfactorily, as shown in fig. 1 C-F.

35. Variations in the early development of the kidney in pig embryos with special reference to the production of anomalies. Frederic T. Lewis, Harvard Medical School, and James W. Papez, Atlanta Medical College. Presented by Professor Papez.

In the summer of 1914, the collection of 165 series of 10 to 12 mm. pig embryos used for class instruction at the Harvard Medical School, was carefully examined to detect anomalies in the region of the developing kidneys. Although fused kidneys of the horse-shoe type are said to occur not infrequently in adult hogs, the proportion of cases is not such that a kidney of this type might be expected in the number of series examined, and none was found. However, the normal relations of the kidneys to one another varied in such a way that we may offer a new explanation of this anomaly, namely that it is due to the relation of the kidneys to the bifurcation of the aorta into the umbilical or common iliac arteries. This bifurcation forms a U-shaped crotch in which the kidneys are lodged, and from which they escape by migrating upward. The arteries, as a mechanical obstruction, tend to bring the right and left renal blastemas close together, so that fusion may readily take place. A fusion at the upper poles, making a horse-shoe kidney convex superiorly, would probably arise earlier than the fusion at the lower poles, in which case the horse-shoe would be convex inferiorly. The relations of the kidneys which seem to justify this interpretation have been demonstrated in a series of models.

The anomalies actually observed consist chiefly of diverticula of the Wolffian duct, perhaps representing abortive ureters. Eighteen of these were found, most of which are on the part of the Wolffian duct

distal to the orifice of the ureter. One is detached, forming an epithelial cyst. Two diverticula were found springing from the ureter itself. One of these which is slightly elongated, ending blindly near the renal blastema, might give rise to a divided ureter, but inasmuch as it does not enter the blastema, no renal tubules would empty into it. On the proximal side of the orifice of the ureter in the Wolffian duct, there were six diverticula, generally close to the ureter. Thus a portion of the Wolffian duct immediately below the lowest and typically rudimentary tubules of the Wolffian body is generally free from diverticula. In one case, however, an elongated diverticulum in this region extended toward the Wolffian body and ended in relation with a blastemal cyst, such as produces the glomerular end of a renal tubule. In position and structure this formation is intermediate between the Wolffian body and kidney. In the anterior end of the Wolffian body, the elongating cysts open directly into the Wolffian duct, but below, as is well shown in this instance, the Wolffian duct sends out tubules, comparable with the ureter and collecting tubules of the kidney, to join with the portion of the tubule derived from the cyst. This specimen shows also a second and much smaller outgrowth of the Wolffian duct, nearer the ureter, which brings the Wolffian body into still closer relation with the permanent kidney.

36. Some anatomical deductions from a pathological temporo-mandibular articulation. FREDERIC POMEROY LORD, Department of Anatomy and Histology, Dartmouth Medical School.

In a previous paper, "Observations on the temporo-mandibular articulation," certain reasons, based largely on the study of a working model of the jaw-joint, were given to prove that the mouth is opened by the combined pull of the external pterygoid muscles. It was also shown that, during opening, each condyle of the jaw moved forward nearly in a straight line, which is almost parallel to the plane of pull of the two external pterygoid muscles; and that the depth of the bony glenoid fossa is practically obliterated by the inter-articular cartilage.

Further proof of these facts has been noted in a singular skull, found in the Peabody Museum at Harvard, whose Director, Professor Putnam, kindly gave me access to its large collection of skulls.

In this specimen the left jaw-joint is virtually normal; the right has suffered from an attack of osteo-arthritis, apparently recovered from, later. During the attack the whole of the joint surfaces had been remodelled along entirely new lines, without a meniscus, and yet it gave, it would seem, equally good function with that of the other side, representing the usual condition.

The new joint shows the course taken by the advancing condyle, permanently recorded in bone, and the evidence as to the character and direction of the condylar path, thus disclosed, corroborates that previously adduced.

The joint surfaces show, also, better than those of a normal specimen, how well adapted they are to resist lateral or mesial displacement of the

condyles, in closing the mouth, either by the pull of the masseter or the internal pterygoid, and in proper proportion to the direction of their pull.

By making an artificial meniscus, exactly fitted to that especial skull, in the case of a normal specimen, the same adaptation of the joint surfaces can be demonstrated.

37. Distribution of nervus terminalis in man (lantern). Rollo E. McCotter, Department of Anatomy, University of Michigan.

Johnston and Brookover were the first to observe the presence of the nervus terminalis in man. Apparently, the material used by them permitted only of the examination of a portion of its intracranial course. By means of gross dissections of the heads of several human fetuses the writer is able to demonstrate the intracranial course and nasal distribution of the nervus terminalis. The nerve appears on the cortex in the region of the olfactory trigone, courses as a single trunk over the medial surface of the olfactory tract and breaks up into a plexus on the medial surface of the olfactory bulb where it is associated with the vomero-nasal and olfactory nerves. From the plexus on the medial surface of the olfactory bulb the fibers of the nervus terminalis collect into several communicating filaments and course over the lateral surface of the crista galli and pass through the cribriform plate well forward. The nervus-terminalis reaches the nasal cavity as a single bundle and is distributed to the septal mucosa anterior to the path of the vomero-nasal nerves.

This article will be published, with figures, in volume 9 of the Anatomical Record.

38. On the anatomy of the brain and ear of a fish from the coal measures of Kansas. Roy L. Moodie, Department of Anatomy, University of Illinois, Chicago.

The preservation of the soft parts of extinct animals has always been a matter of great interest to students of paleontology and a number of papers have appeared on this subject. There are now known from the studies of various paleontologists muscle and kidney tissues from the Devonian, alimentary canals and muscle tissues from the Carboniferous and from the succeeding formations a variety of the softer organs have been preserved in different ways. They may be mummified, carbonized or changed into mineral substances, or the form of the part may be preserved as a cast of the cavity which the organ occupied. The latter is the usual mode of formation of fossil reptilian and mammalian brains. The casts are, however, always dural casts which never repeat the exact topography of the organ, and the smaller convolutions of the brain are not represented in the average brain cast.

A study of the brain cast of a mammal would give a more accurate idea of the form of the brain than would the cast of the brain case of a reptile, since the mammalian brain more nearly fills its cavity than does the brain of a reptile, as noted by Dendy (Phil. Trans., Royal Soc.

London, Ser. B, vol. 291, pp. 227–331) for Sphenodon. The brain case of all recent selachians and teleosts is much larger in proportion to the size of the brain than among the reptiles, so that a cast of the cranial cavity of a fish would give no idea of the detailed anatomy of the brain. We may be sure that the organs which are described herewith are not casts but are, apparently, a transformation of brain substance, before decomposition, into some mineral, probably calcium phosphate. The walls of the brain are not shrunken but preserve a rounded contour as they probably had in life, resembling greatly, so far as details are concerned, the brain of a recently dissected and well-preserved fish. There are also preserved in their proper relations nerves and blood vessels, somewhat enlarged by the segregation of mineral matter and the subsequent formation of crystals but still preserving the normal relations. It is hard to conceive of this method of replacement of the brain by mineral matter in view of the chemical analysis of the brain which shows such a high percentage of water and soluble substances, and such a small percentage of resistant substances such as neurokeratin.

The little fossils with which we are at present concerned were collected in shales above the Kickapoo limestone in the Coal Measures near Lawrence, Kansas. The nodules containing the brains are all small, the specimens of brains themselves measuring only 15 mm. in length. The skeletal parts of the fishes have largely disappeared, so far in fact that it is not possible to determine the nature of the skull. Identification of the form being thus impossible, we are forced to use the characters of the brain to locate our form. Fortunately for this purpose Eastman has described a small brain from the Waverly of Kentucky, Iowa Geol. Surv., vol. 18, 1908, p. 267, pl. 13, very similar in many ways to the brains from Kansas. He was fortunate enough to identify the species of the fish to which the brain belonged, naming it Rhadinichthys deani and placing it among the Chondrostei or ganoids. The fish described by Eastman was collected in the Mississippian of Kentucky, but the character of the brain is so similar to those from the Coal Measures of Kansas that we will be quite safe in locating our fish near the Chondrostei, to which certain characters of the brain ally it independent of any comparison.

The brain itself is very completely shown in a series of specimens and we are able to study all sides of the brain in a few cases. The spinal cord is only partly represented, if at all, by a very small portion on the edge of the nodules. The vagal lobe is single and lies far back over the region of the fourth ventricle. There are no indications of separation of the lobe into subdivisions as is so common among existing fishes. Its complete form is preserved but the one best preserved has the upper surface abraded, since it projected slightly through the surface of the nodule. The facial lobe is smaller in the fish from Kansas than it is in the Mississippian brain described by Eastman. It is separated from the vagal and cerebellar lobes by slight constrictions and from its anterior aspect there arises a tubular structure which is apparently connected with the pineal organ. This may be a vessel of some description or it

may be a fold of membrane which has been preserved. The cerebellar lobes are most unusual in being entirely lateral. If the median portion of this organ has become involuted below the surface of the huge optic lobes it is not possible to determine this. A study of the internal construction of the brain is not possible with the material at hand, if it will ever be; the formation of large crystals having obliterated all structural characters. Between the medial tips of the cerebellar lobes lies a structure which may be the pineal body. It is not present in all of the specimens, being entirely absent in one well-preserved brain. From the anterior, ventral portion of this organ runs a rounded elevation which may be either the stalk of the epiphysis or a plexiform vessel. Its morphology is uncertain. This is a very constant structure among the specimens at hand. The optic lobes are very large and indicate, apparently, a teleostean character for the fish. They occupy one-third the full length of the brain and constitute fully one-half its bulk. The eye was large as is indicated by an impression of the orbit and the optic stalk short. The optic chiasm is evident in one specimen but the details of its nature are uncertain. The structure just anterior to the optic lobes is probably the thalamus or praethalamus. It, like the vagal lobe, has its dorsal surface somewhat abraded but other specimens show that its full form was not greatly different from that shown in the figures. The olfactory lobes are distinct and relatively large, being separated by a slight groove. The base of the olfactory tract is preserved and shows a strong olfactory development. The horizontal semicircular canal is well preserved on the right side of one specimen and on both sides of another nodule. The ampulla is large and the utriculus nearly double its size. The base of the vertical semicircular canal is preserved on the upper aspect of the utriculus. The base of the hypophysis is large and well preserved.

My thanks are due Doctor Herrick and Doctor Johnston for assistance in the determination of the characters of this little Paleozoic brain. A fuller discussion with illustrations and a review of other fossil brains will appear shortly in the Journal of Comparative Neurology.

39. The growth of the vascular system as it is correlated with the development of function in the embryos of amblystoma. JULIA S. MOORE, (introduced by GEORGE E. COGHILL).

Embryos of Amblystoma punctatum and microstomum in the physiological stages of development described by Coghill (Jour. Comp. Neur., vol. 24, p. 163) as (1) non-motile, (2) early flexure, (3) coiled-reaction, (4) early swimming stage, were used in this study of the vascular system in its correlation with other organ systems and with the growth of the embryo as a physiological unit. The following correlations are made upon the basis of a close study of serial sections and of living embryos.

As in other vertebrate embryos, rhythmic contractions of the heart begin before there is any connection with the nervous system and before there is any histological evidence of the muscular nature of the myocardium. The first cardiac movements do not occur until after

the body movements, as represented by the early flexure stage, are well established. The rate of heart-beat averages in the early flexure stage 29 per minute; in the coiled-reaction stage, 49. In the early swimming stage it varied from 49 to 72. At the time that rhythmical contraction begins there is no perceptible connection between the arteries and the veins. In the coiled-reaction stage communication is established between the branchial vessels of the first gill and between the internal carotid artery and the ophthalmic vein, and a circulation of plasma may begin at this time. But corpuscles in living embryos were seen to circulate first in the late coiled-reaction or the early swimming stage, though they are found in the heart and venous system earlier in the microscopic sections. It is evident that circulation of corpuscles in the gills is started about the time that definite swimming begins, circulation in the balancer follows very soon afterwards. The evidence obtained concerning the aerating function of the corpuscles is not conclusive. From all observations thus far made it would seem that the corpuscles carry no haemoglobin until a later period than that considered in this paper.

Up to the early swimming stage no blood vessels could be seen to enter the myotomes, and no vascular connection has been made with the digestive system, which is differentiated even less than the myotomes at this time. Circulation could be seen in the vessels between the myotomes only at a later period. The mouth of the embryo does not open until some two weeks after swimming begins. At the non motile stage the cells of the ectoderm and the nervous system already show a greater degree of differentiation than those of other organ systems, as signified by the diminution of the yolk content of the cells. A similar diminution of yolk is marked in the blood corpuscle about the time that it comes into circulation. By the time that the corpuscles have used up their yolk they have approximately attained their adult size and form. The cells of the pronephros show a similar differentiation and diminution of yolk about the same time or a little earlier. The entodermal and mesodermal cells are still crowded with yolk up to a later period. A characteristic relation seems to exist in all cells between the nuclei and the yolk, which suggests a process of digestion within the cell. The relative amount of yolk thus seems to hold a definite relation to the degree of differentiation in the various parts of the embryo. The facts observed in connection with the rate of differentiation and the disappearance of yolk in the various parts of the embryo would indicate that the blood performs little or no nutritive function, as each cell of the body seems to be able to supply its own needs both for differentiation and for function, until swimming is well established.

In the early swimming stage no blood vessels are yet found in the nervous system, but the anterior cerebral vein is very closely applied to the surface of the brain, while the segmental vessels, developing later in the trunk come into close contact with the spinal cord. This close relation in development of the anterior cerebral vein with the most highly differentiated parts of the brain would indicate a correlation of the

vascular system with the early processes of function and differentiation in the nervous system. The tetanic condition of the embryo in the coiled-reaction necessitates a violent metabolism whose products must be removed. The nerve centers controlling this muscular activity must also undergo considerable metabolism. Hence it may be supposed that the early differentiation and function of these parts have stimulated the development of the vascular system, and that the vascular system has an excretory function in relation to these parts. The presence of the sacculated outgrowth of the dorsal aorta at the level of the pronephros, the ciliated nephrostome, the opening of the pronephric duct into the cloaca, and the close relation of the posterior cardinal vein to the pronephros indicate that the vascular system in conjunction with the pronephros is functional as an excretory system in the coiled-reaction stage. The communication of afferent and efferent vessels in the gills, permitting a circulation of plasma, makes possible the excretion of carbon dioxide through the blood in the coiled-reaction stage, while a distinctive aerating function can appear only later with the development of haemoglobin.

40. A preliminary note on the septum secundum in the pig. C. V. Morrill, Department of Anatomy, University and Bellevue Hospital, Medical College.

In the course of a paper devoted chiefly to the development of the Purkinje fibers of the heart, Retzer (some results of recent investigations on the mammalian heart; Anat. Rec., vol. 2, no. 4, 1908) briefly discusses the formation of the atrial septum in the pig. He states that the accounts of His and Born though accepted by most embryologists are incorrect on this point. In the pig Retzer considers that septum II in Born's sense does not exist and that this supposed septum is merely a fold in the atrial wall produced by the growth of the auricles around the conus arteriosus as a fixed point; and further that it never attains sufficient size to justify its being called a septum.

Since, as Retzer says, Born's account has been followed to a large extent by other writers as evidenced by the descriptions of Hochstetter in Hertwig's Handbuch and Tandler in Keibel and Mall's Manual, it seemed worth while to re-examine the development of the atrial septum in the light of Retzer's criticism.

The study of this point is based on serial sections of pig embryos of 6.8, 7.9, 8.5, 12.3, 15.2 and 21.0 mm. total length. Of these, the heart regions of the 7.9 and 15.2 mm. embryos have been reconstructed in wax and that of the 21.0 mm. is in process of reconstruction.

In the 6.8 mm. stage, the earliest examined, septum I forms an incomplete interatrial curtain. Both ostia are present. The caudal (inferior) border of septum I meets and fuses with the corresponding wall of the atria. At the ventral end of the line of fusion, a slight thickening projects into the right atrium and with this the caudal extremity of the left sinus valve blends. In the 7.9 mm. embryo, septum I has fused with the endocardial cushions for the most part,

but a narrow slit, the remains of ostium I, still connects the two atrial chambers. Ostium II has enlarged and its borders are fimbriated. The thickening in the caudal wall of the right atrium close to the ventral extremity of septum I, which was noticed in the earlier embryo, has developed into a distinct spur which extends cephalad (upward) a short distance in the ventral border of septum I, just dorsal to the still-persisting ostium I. This, I believe, represents the earliest appearance of the septum II of Born (he did not describe the earlier stages). With it, the caudal end of the left sinus valve blends. The corresponding end of the right sinus valve is lost in the caudal wall of the right atrium, close to this point. In the 12.3 mm. embryo, the spur has thickened and extends further cephalad. In the 15.2 mm. stage, it has lengthened out into a definite ridge extending from its place of origin in the caudal wall of the right atrium, first cephalad, then dorsally, arching over ostium II to reach the dorsal atrial wall, where it fades out. Its caudal end is thick and up-standing; its cephalic end narrow, pointed and not sharply marked off from the atrial wall. The caudal ends of both sinus valves are now fused with it. In this stage ostium I has entirely closed and ostium II considerably enlarged dorso-ventrally, so that the free border of septum I bordering the latter opening, now faces almost entirely cephalad (upward). In the 21.0 mm. stage, the oldest examined, septum II has become thicker and more sharply defined near its caudal extremity. Its narrow, pointed end extends cephalad and dorsally, bordering ostium II, then caudally for some distance along the dorsal wall of the right atrium in the region of the spatium intersepto-valvulare and close to the left sinus valve.

It does not seem probable that this very definite thickening is merely a fold in the atrial wall produced by the growth of the auricles around the conus, as Retzer claims. It is true that in the middle of its course it does conform to the curve of the conus, but at its caudal extremity where it is thickest and at its pointed extremity which lies in the dorsal atrial wall, it is entirely unrelated to that structure. Thyng (the anatomy of a 17.8 mm. human embryo; Am. Jour. Anat., vol. 17, no. 1, 1914), has recently recorded the presence of a 'ridge or tubercle' in the caudal part of the right atrium which he considers to be the caudal end of the future septum secundum in the human heart.

A more detailed account of this structure and nearly related parts will be given in a subsequent paper.

41. Studies on the syrinx of Gallus domesticus. J. A. Myers, Institute of Anatomy, University of Minnesota, Minneapolis.

The results of this work may be summarized as follows:

Structure. (1) The syrinx of the domestic chicken belongs to the tracheo-bronchialis type, and is quite simple when compared with the voice organ of song birds. (2) No intrinsic muscles are present in the syrinx of Gallus domesticus. The extrinsic paired sterno-trachealis with its caudal prolongations constitute the entire musculature of the syrinx. (3) The rigid skeleton is very highly modified. The first

four tracheal rings are imperfectly fused to form the tympanum. The four intermediate syringeal cartilages are continuous ventrally with the ventral pyramid of the pessulus, while dorsally they end unattached. The first bronchial half-rings are large and in adults are attached and fused at both ends of the pessulus. The pessulus is the largest of all skeletal parts and lies dorso-ventrally at the junction of the bronchi in a plane transverse to the long axis of the trachea. The tracheal rings, the pessulus, and the ventral ends of the first half-rings become ossified, while all other skeletal parts remain cartilaginous. (4) The external tympanic membranes appear between the fourth intermediate syringeal cartilages and the first half-rings while the internal tympanic membranes extend from the caudal borders of the pessulus to the bronchidesmus and represent merely a modified part of the medial bronchial walls. (5) The syrinx is lined with stratified ciliated columnar epithelium containing numerous simple alveolar glands. Upon approaching the tympanic membranes this columnar epithelium is transformed into a stratified squamous epithelium which becomes a single layer of flattened cells over the membranes proper. (6) The tympanum is attached to the remainder of the syrinx only by elastic membranes.

Development. (1) The first indication of the respiratory system was observed in a 68 hour embryo in which the laryngeo-tracheal groove and the bronchi were represented. At first the trachea is much shorter than the bronchi, but with the development of the neck, it becomes, after the 140 hour stage, relatively much longer than the bronchi. The walls of the trachea and the bronchi are at first composed only of epithelium which contains two or three rows of nuclei. (2) The mesenchymal condensation common to the entire epithelial tube first becomes markedly prominent at the tracheal bifurcation in an embryo of 152 hours. (3) The anlagen of the first bronchial half-rings appear in a 176 hour embryo, those of the fourth intermediate syringeal cartilages appear 12 hours later. The anlagen of the third intermediate syringeal cartilages and the anlage of the pessulus are present at 200 hours. (4) Distinct cartilage cells were first observed in the first bronchial half-rings. (5) The first four tracheal rings have not united to form the tympanum at hatching, nor have the other skeletal elements taken the shape of those found in the adult. No bone is present at the time of hatching. (6) Ciliated cells are present in stages beyond 248 hours but were not observed in the region of the future tympanic membranes. (7) Mucous cells were first observed in 332 hour embryos and only in later stages were they found arranged in the form of simple alveolar glands. (8) Muscular tissue is differentiated in the 176 hour stage. Muscle fibers showing faint cross striations appear at 296 hours. At 452 hours the muscles are well developed. (9) At the time of hatching the tympanic membranes are thick. They are covered, however, as in the adult, with a single layer of epithelial cells.

Function. (1) That the syrinx is the true voice organ of the chicken is evident from the following deductions: First, structurally it is the only part of the respiratory tract capable of producing sound; Second,

when the trachea is divided and the cephalic portion tightly tied off, the chicken is still able to crow; Third, after division of the trachea, voice can be reproduced artificially by forcing air into the air sacs. (2) The upper larynx serves only to modulate the voice. (3) The sterno-tracheal muscles by their contraction shorten the trachea and modify pitch. They also draw the tympanum cephalad, thus indirectly varying the tenseness of the tympanic membranes. (4) The air sacs are necessary in voice production, for voice could not be reproduced artificially after puncturing the cervical sacs.

42. On the presence of elastic ligaments in the middle ear region of birds. A. G. Pohlman, St. Louis University.

Recent work by otologists, notably Kreidl and Mangold, has demonstrated that our knowledge of the anatomy and physiology of the middle ear region is imperfect. The Tensor tympani and Stapedius muscles are regarded as synergists rather than opponents and even the facial nerve innervation to the Stapedius is denied on an experimental basis. The problem of what these two muscles really work against is of interest not only to the physiologists but to the anatomists as well and that they may have some function relative to respiratory and atmospheric changes in the air content of the middle ear is not to be denied. The bird because of its single columella and single Tensor tympani presents a condition where the physiology may be more easily determined but Denker's work on the parrot does not consider the details of the middle ear and Breuer's article, while it takes into account the functions of the single muscle, quite avoids all mention of the ligamentous apparatus. The most recent work on ligaments is that by Smith who describes the position of Platner's ligament and the two accessory drum ligaments in the chicken quite accurately.

It was assumed that the Tensor tympani in birds must pull against elastic ligaments, and the following points were developed: (1) That the attachment of the stapedial plate to the oval window and the membrane of the Fenestra cochleae were elastic in nature. (2) That Platner's ligament, placed in direct opposition to the pull of the Tensor tympani, is also elastic and draws the columella forward to its position of rest when the muscle relaxes. (3) That the drum attachment itself is rich in elastic fibers. (4) That in some birds elastic fibers and even ligaments reach from the extra-columella over the drum into the Eustachian tube. The conditions in the mammal remain to be investigated.

43. A genetic interpretation of the stapes, based on a study of avian embryos in which the development of the cartilaginous otic capsules has been experimentally inhibited. Franklin P. Reagan, Department of Comparative Anatomy, Princeton University. (Introduced by C. F. W. McClure.)

The chondro-crania of all vertebrate embryos possess one essential ground plan. Around the anterior end of the notochord, is formed the parachordal cartilage, anterior to this the trabeculae. Following the

formation (invagination) of the epithelia of the three bilaterally symmetrical sense organs, the latter become more or less surrounded by prechondral cytoblastemae, later by cartilages, that surrounding the otocyst being the anlage of the otic capsule or cartilaginous labyrinth.

It occurred to the writer that this cartilage of the otic capsule evidently arises in response to the presence of the auditory epithelium, and that if this be true, an excision of the epithelium at an early stage would inhibit the stimulus to the development of an otic capsule, and that further, if this also be true, it would be possible to test to what extent the stapes can develop in the absence of a cartilaginous otic capsule, or in the absence of the stimulus which produces the latter.

Fortunately it was found that the removal of the otocyst from one side of chick embryos from five to sixty hours incubation resulted in the complete inhibition of the development of the cartilaginous otic capsules, as revealed by a study of operated embryos which were allowed to incubate from six to fifteen days.

On the operated side, the staff-like portion of the stapes resembles in shape, size and position the same portion on the uninjured side, seemingly complete except lacking the flange-like ring of cartilage which completes the stapedial plate.

It seems that the central portion of the stapedial plate is actually formed by cartilage of the hyoid arch while its periphery arises as an independent chondrification in the fenestra ovalis, distinct from the otic capsule but incapable of developing under conditions in which the latter fails to form. Both the otic capsule and the periphery of stapedial plate appear to have as their exciting stimulus the auditory epithelium.

I have reason to believe that my evidence is not merely of a negative sort, resulting from injury to the mesenchymatous anlage of the otic capsule.

44. On the origin of the duct of Cuvier and the cardinal veins. Florence R. Sabin, Anatomical Laboratory, Johns Hopkins University.

Certain injections of young embryonic pigs brought out clearly the fact that the posterior and mesial or sub-cardinal veins arise as longitudinal anastomoses of direct lateral branches of the aorta. These lateral branches are distinct from the dorsal segmental branches which pass directly to the spinal cord. The lateral branches on the other hand alternate with the nephritic tubules and are two or three to a segment according to the number of tubules. The mesial cardinal vein in the pig forms at the same time as the posterior cardinal, but it is the posterior cardinal vein which connects with the duct of Cuvier. The specimens of injected pigs will be demonstrated at this meeting.

These studies in the pig led to taking up the subject of the origin of the cardinal veins in the chick since the material is so much easier controlled. In the chick of 36 hours incubation I injected a little India ink into the marginal vein and watched it circulate through the embryo. All of the ink simply passed through the heart and the double aorta back into the capillaries of the membranes so that there was no capil-

lary circulation within the embryo. In chicks a little older, however, from 38 to 42 hours of incubation I saw a little of the ink pass through a tiny branch from the lateral surface of the aorta opposite the venous end of the heart around to the vitelline veins on either side. As soon as this connection between the aorta and the venous end of the heart is established the embryo itself as distinct from the membranes may be said to have a circulation. This tiny vessel which is the first part of the duct of Cuvier to develop grows from the aorta just cephalic to the first cervical myotome. Subsequently direct branches from the aorta opposite about three segments grow around to the vitelline veins and these primitive branches at once show anastomoses.

At the same time that this venous return for the blood of the embryo is formed, sprouts grow to the brain from the arch of the aorta. In the early chick the arch of the aorta is just at the root of the optic vesicle and it is at this point that the brain first receives capillaries. At first these capillaries have no circulation since they have no connection with the venous end of the heart. Gradually the capillaries to the brain spread out around the base of the optic cup and over the mid-brain. From this plexus a single vessel grows caudalward along the side of the medulla ventral to the otic vesicle and just at the caudal end of the medulla turns sharply ventralward almost at a right angle and joins the cephalic part of the duct of Cuvier. I have one specimen in which the branch of the duct of Cuvier connects both with the aorta and with this vessel from the brain. Soon, however, the direct connection between the aorta and the heart is broken and the primitive head vein is established. Thus the composite nature of the head vein, suggested by Salzer in 1895 and more fully described by Grosser in 1907, is explained. The part of the head vein which lies close to the neural tube arises from the arch of the aorta and is a part of the vascular system of the central nervous system; the caudal part of the head vein arises directly from the aorta just cephalic to the first cervical myotome, it lies in the lateral groove and is analogous to the posterior cardinal vein. The part of the head vein cephalic to the first myotome may well be called the vena capitis as was done by Grosser while the caudal part which becomes the internal jugular vein is a true anterior cardinal vein in the sense of being analogous to the posterior cardinal vein. This caudal part of the vein is much the shorter portion of the head vein in the early chick. It should be noted that it is the entire head vein which is usually termed the anterior cardinal vein. In this use of the name it must be emphasized that the head vein has a double origin.

The posterior cardinal vein in the chick is likewise formed from direct branches of the aorta which grow lateralward to the Wolffian body and there form a longitudinal vein. In contrast to the pig these branches are for the most part segmental arteries that is they arise between the myotomes. A few lateral branches opposite the myotomes are involved in the formation of the posterior cardinal veins in the chick but they are always less numerous than the segmental vessels. The position of origin of the segmental vessels along the aortic wall varies

in the different segments and seems to follow a line corresponding to the lateral surface of the spinal cord, but the vessels grow directly lateralward to the groove just ventral to the myotomes. Moreover, when the nephritic tubule is present the longitudinal vein lies closer to it than to the myotome. The difference in the development of the posterior cardinal vein in the pig and in the chick seems to correspond to a relative difference in the time of development of the nephritic tubules.

Thus to sum up the origin of the venous system, the first vein of the embryo is the duct of Cuvier which is a direct connection between the aorta and the vitelline veins. The cardinal system in general arises as a longitudinal system of veins from direct branches of the aorta. The cardinal system proper extends throughout the zone of the myotomes and lies in the Wolffian groove ventral to the myotomes. In the chick the direct connection between the aorta and the heart occupies the zone of the first three or four myotomes. Eventually the plexus of vessels representing the duct of Cuvier in the chick covers the zone of the first seven myotomes. In the pig the posterior cardinal vein develops from lateral branches at the same time as the nephritic tubules and these lateral branches alternate with the nephritic tubules. In the chick the posterior cardinal vein develops more rapidly than the tubules and comes in part from lateral branches of the aorta which are intersegmental but mainly from dorsal segmental branches which however do not grow first to the spinal cord but rather directly lateralward to the Wolffian groove where they anastomose to make a longitudinal vein.

45. The technique of Weber's method of reconstruction. Richard E. Scammon, Institute of Anatomy, University of Minnesota. (Lantern slides.)

This method as used by its author was applied mainly to surfaces which were almost flat. Its extension to the study of rounded surfaces requires that a method of curvature elimination be adopted. In dealing with the surfaces of cylinders or cones this is easily done by correcting for vertical curvature alone, but where segments of spherical surfaces are involved corrections for horizontal curvature are also necessary. Vertical and horizontal corrections can be made so that the finished plat will give an approximately true representation of both the area and shape of a given outline upon a curved surface.

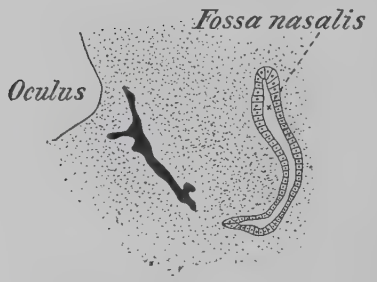
The method of making a finished reconstruction from the reconstruction plat has been considerably simplified by the introduction of color by means of 'Herring' papers and by building up the final reconstruction in strata.

The entire process involves the following steps: (a) Correcting section drawings for vertical curvature; (b) Scaling drawings for thickness; (c) Laying out reconstruction plat with corrections for vertical and horizontal curvature; (d) Plotting gauge lines; (e) Building up the finished reconstruction from the plat.

46. Nasolacrimal duct diverticula and their genetic significance (a preliminary note). J. Parsons Schaeffer, The Daniel Baugh Institute of Anatomy and Biology of the Jefferson Medical College.

It is generally believed that the wall of the nasolacrimal duct are regular and that the lumen of the duct represents a more or less uniform cylinder. Indeed, this is what one gathers from many text-books and from the average gross dissection of the channel. The common practice of passing the lacrimal probe would also lead one to think that the nasolacrimal duct has even or regular walls.

1



2

3

Fig. 1 Outlines of the lumen of the ductus nasolacrimalis at various levels. What appears to be two ductus nasolacrimales lying side by side at one level turns out to be the main duct and a diverticulum from it. From an adult.

Fig 2 Frontal section through the nasal fossa of a forty-day human embryo. The anlage of the nasolacrimal passages is indicated in solid black. Note its complete isolation from its former surface connection. Note a few lateral buds from the mother cord of cells, presumably the proton of diverticula. At the ocular end of the cord the lacrimal ducts are beginning to sprout.

Fig. 3 Showing the irregular canalization of the ductus nasolacrimalis, from a term child.

Admittedly, such a condition does obtain in a certain percentage of cases and represents one of the anatomic types of the nasolacrimal duct (fig. 4). On the other hand, recent investigations by the writer indicate that many nasolacrimal ducts present lumina of very irregular contour, some even more or less tortuous in course. The irregularity and complexity of the lumen of the nasolacrimal duct is at times carried to a marked degree (fig. 4).

Minor irregularities are at times due to mere folds in the mucous membrane. In many instances, they are of little moment, again, they may form definite bridges along the walls of the duct.

In some instances two parts of the nasolacrimal duct are not in exact line and the connection between the parts a somewhat deviating cross-channel. Finally, many nasolacrimal ducts present very irregular lumina due to diverticula.

These diverticula vary from those of an insignificant size to those with relatively large dimensions. The diverticula are obviously direct extensions of the walls of the duct proper. They are lined with a mucosa

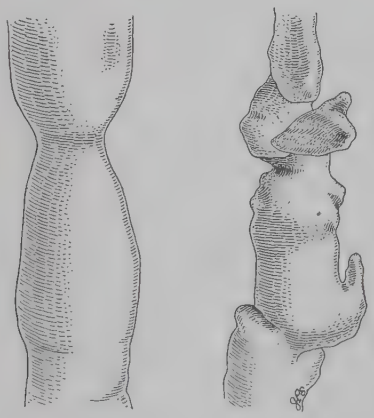


Fig. 4 Showing outline of reconstructions of lumina of mid-portion of two adult nasolacrimal ducts, one is very regular, the other every irregular and with diverticula.

similar to that lining the main duct, and at the ostia of the diverticula the mucosae of the main duct and diverticula are directly continuous, both grossly and histologically.

In studying cross-sections of the nasolacrimal duct, one is at times puzzled to explain what are apparently two ducts lying side by side. However, by following the sections serially one finds that the one cavity sooner or later communicates with the other, i. e., one turns out to be the nasolacrimal duct proper and the other merely a diverticulum from it (fig. 1).

These diverticula must be very important clinically and they need further study. Owing to the irregularity of the lumen in many instances of the nasolacrimal duct, it is obvious that false passageways are repeatedly made by operators when they pass the lacrimal probe.

Genetically, nasolacrimal duct diverticula are doubtless the result of irregular canalization in fetal life of the solid cord of epithelial cells from

which the several nasolacrimal channels develop. One must, therefore, return to the embryologic stage of the nasolacrimal duct for a proper genetic interpretation of the nasolacrimal duct diverticula.

After the strand of thickened epithelium (the anlage of the nasolacrimal passages) along the floor of the now rudimentary naso-optic groove becomes entirely isolated from its surface connection it becomes entirely surrounded by mesenchymal tissue and is for a time without a lumen (fig. 2).

This epithelial cord becomes canalized in a very irregular manner. In this canalization, one has direct evidence of the earliest stages of nasolacrimal duct diverticula. Small lateral pouchings from the main channel, due to a re-arrangement of epithelial cells, are early in evidence. *Para passu* with the growth of the main duct, the diverticula increase in size. At times one finds direct side branches from the mother cord of epithelial cells and some of them doubtless represent the proton of diverticula (fig. 2).

It must, therefore, be concluded from the evidence at hand at present that the diverticula from the ductus nasolacrimalis are of congenital origin and are not acquired in later life.

47. On the gross morphology, topographical relations, and innervation of the human parotid gland. S. S. SCHOCHET, Department of Anatomy, Tulane University. (Introduced by Irving Hardesty.)

The parotid gland hardened *in situ* presents an irregular three-sided inverted pyramid with base uppermost and apex below. The posterior surface of the gland presents a marked concavity with three depressions: an external, middle, and an internal concavity. In addition there are in the inferior portion of the mesial surface three grooves caused by the styloid process, the diagastric muscle, and the sterno-cleido-mastoid respectively.

The external carotid artery was not found buried in the gland substance in any of the six specimens so far examined, and the temporomandibular vein lies in a more external and deeper plane. These relations differ from the descriptions given in all the standard text-books. In the illustrations of Testut these vessels are represented as buried in the gland substance.

A small oval or fusiform thickening of irregular outline of a yellowish white color has been found embedded in the auriculo-temporal nerve. Sections of this show it to contain numerous ganglion cells. Because of the position and the branches of connection of this body, it is here named the "parotid ganglion" (ganglionun parotidis). It measures from 1 to 1.5 cm. in length and from .25 to 0.5 cm. in thickness. It is supported by considerable fibrous connective tissue. It is located a short distance from a point where the two roots of the auriculo-temporal nerve fuse after encircling the middle meningeal artery, and in close relation with the temporo-mandibular articulation, the internal axillary and temporal branches of the external carotid artery, thus lying in the plane of the posterior mesial surface of the parotid gland. It should be num-

bered among the sympathetic ganglia of the head, and included, with the ciliary, spheno-palatine, otic, and sub-maxillary ganglia, as a ganglion of the cephalic sympathetic plexus.

The branches of distribution of this ganglion are mainly through the parotid branches of the auriculo-temporal nerve. This ganglion is assumed to serve as a common point of termination of visceral efferent axones from the glosso-palatine nerve (nervus intermedius) which axones reach it by way of the small superficial petrosal nerve, and pass uninterrupted, through the otic ganglion to terminate about its cells; these cells in turn send their axones into the substance of the parotid gland. It is also possible that some of the visceral efferent axones of the glosso-palatine are incorporated in that part of the facial nerve which passes to the parotid ganglion by the two communicating branches to the auriculo-temporal nerve.

Serial stained sections of the entire human parotid gland are examined to determine the presence of other sympathetic ganglion cells in its capsule and gland substance.

48. Comparative study of certain cranial sutures in the Primates. R. W. Shufeldt, Washington, D. C.

Early in February, 1914, Doctor Ales Hrdlicka, in charge of the Department of Physical Anthropology of the United States National Museum, invited my attention to certain variations and peculiarities to be found in the sutures on the lateral aspect of the skull in man, and suggested that I should examine into the matter, with the view of publishing a report upon my subsequent studies of the subject. Doctor Hrdlicka very kindly gave me every facility to examine, compare, and photograph the enormous collection of human skulls of which he has charge at the Museum, or such of them as I intended to employ in my investigations.

Hardly had I gotten into these researches when I came to the conclusion that my work would be a far more useful contribution to anthropology were I to include in it a similar series of comparisons made with the skulls of various species and genera of apes, monkeys, marmosets, and their allies. To this end I applied to Mr. Gerrit S. Miller, Jr., Curator of the Division of Mammals of the United States National Museum, who kindly permitted me to examine the superb collection of the skulls of these animals under his charge at the Museum, and to make such photographs as I required for illustrations. In this matter I was very materially assisted by Mr. Ned Hollister, Mr. Miller's aid at the Museum. To all three of these gentlemen I am under great obligations and I have pleasure in thanking them for their assistance in my work, without which it would have been entirely impossible for me to have taken it up in any satisfactory manner whatever. Data from one or two human skulls in my own collection are included in this work, as well as those from skulls of certain monkeys and tamarins, presented me by Mr. Edward S. Schmid, of Washington, D. C.

The locations of the sutures in the human skulls have been known for a great length of time, as have also the bones between which, in any particular case, they may occur. These sutures have, further, all received names which have been bestowed upon them by the older anatomists, and the majority of these names are still employed in our present-day works on human anatomy.

In examining large series of skulls, it will be found that some of their sutures vary but little, as for example, most of those found between the bones of the face; while on the other hand, a very considerable amount of variation is to be observed in some of the cranial sutures, and especially those at the lateral aspect of the cranium. These sutures are caused to vary in accordance with the mode of articulation of certain of the cranial bones, which later, in their turn, present various differences in their articulations that are responsible for those sutural variations. Again, as is well known, certain sutures present variations which are due to the presence of certain supernumerary or epactal ossifications which are intercalated between the cranial bones in the lines of their sutures, examples of which are the Wormian segments and the epipterics—the former usually occurring in the lambdoid suture connecting the parietal and occipital, as well as in the sutures between the parietals and other bones. On the other hand, the adventitious epipterics occur in the spaces of the lateral fontanelles, and are subject to marked variations with respect to number, size and position.

It was these epipterics and the sutures among the bones at the lateral aspects of the cranium, as they are found to vary in the various races of man and in the different families, genera and species of the Quadrumana, that I gave my especial attention, and to which my contribution to the subject is devoted, of which this paper is a brief abstract.

In the course of my work I compared several thousand skulls of men, women, and children of all ages, except the very young. The vast majority of these were of prehistoric Peruvians collected by Dr. Hrdlicka, in addition to which there were a sufficient number from other races of the world to satisfy me that I had obtained in my researches all the known sutural variations worthy of record in the aforesaid region of the cranium, and that variations presented on the part of the epipterics were practically endless in nearly every respect. I made a large number of sketches to show this, the majority of which will appear in my paper when it is published. I also made thirty-six photographs of the lateral aspects of skulls of men, women and children, and of various species of the Quadrumana. These, for the most part, are more than half natural size, and show most interesting variations of the sutures at the two sides of the cranium in the Primates, and, taken in connection with my other data, probably present the most extensive study of these sutures and epipterics up to the present time.

Thousands of figures of photographs of the lateral aspect of the human skull, of both sexes and all ages, have been published, and thousands of descriptions of the sutures in that region of the cranium have ap-

peared. There have also been an enormous number of similar illustrations and descriptions given in works upon comparative anatomy for the Quadrumana. In the vast majority of these figures and descriptions the fact is pointed out that the normal articulatory arrangement of the frontal, parietal, temporal (squamous portion) and sphenoid (greater wing) is, apart from the facial articulations, that both the frontal and parietal bones, separated as they are by the coronal suture, articulate in nearly equal proportions with the superior margin or border of the greater wing of the sphenoid, where we find the spheno-parietal and spheno-frontal sutures; that the squamo-parietal suture is the bounding line between the temporal and parietal, and, lastly, that the squamo-sphenoidal suture occurs between the sphenoid and squamous portion of the temporal. These are the only sutures to be considered here and indicate the remaining articulations.

These articulations may or may not be the same on the two sides of the same skull—indeed, they may exhibit very considerable variation. Moreover, they are to be found in the skulls of all Primates, irrespective of various forms of disease, as hydrocephalus, or in distorted skulls, whether the distortion be congenital or induced.

Craniologists long ago bestowed names on some of the principal points of meeting of certain of these sutures, or where they cross prominent ridges, as the stephanion, where the coronal suture crosses the temporal bridge, and pterion, the point where the temporal, frontal, parietal and greater wing of the sphenoid either are in contact or approach each other.

As already pointed out, the squamous portion of the temporal fails to meet the frontal through the intervention of the spheno-parietal articulation. This spheno-parietal suture varies greatly in length, and when it is reduced to zero, all four of the above-named bones are in contact. This may occur upon both sides of the cranium or only upon one. It may be designated by the word 'contact,' and it occurs only in a certain percentage of skulls, whether they be normal, abnormal, or pathological. Contact of these four bones also occurs in the crania of the Primates below man; this is shown in one or more of my photographs, and I shall probably meet with others, later on, when this paper is entirely completed.

So far as my studies carry me at present I find, and especially among the higher simians, that among the Quadrumana the sutures, at the lateral aspects of the cranium, have all the variations as they occur in man. The occurrence of epipterics of any size in the skulls of the Quadrumana, however, are rare; indeed, up to the present time, I have failed to meet with any such bones in them beyond those of very minute size.

As to the percentage of the occurrence of contacts in the crania of various races of men, I can at this writing only say that it is very small; and how this percentage compares with a similar percentage, taken from the crania of any genus or family of the Quadrumana, I am not, at this time, prepared to say. For the human species, I have prepared

a great quantity of data on this subject, which can not be well presented in a brief abstract, such as is here submitted.

In man, the true epactal epipterics vary greatly with respect to number, form, size, and position. With respect to their relation to the pterion, these epipterics, on one or both sides, may be anterior or posterior, or they may be, as I found them to be in the case of an adult male Cinco Cerros Indian (Peru), anterior, posterior, superior, and middle of the left side, while they were multiple also on the right side, but some of the pieces were here lost. An epipteric may be co-extensive with the pterion, in which case it is total. Sometimes the two sides of cranium agree in these respects—that is, there may be an anterior or a posterior epipteric on both aspects of the skull, while they *never* exactly agree in the matters of form and size. Some epipterics are nearly round, some are triangular, others squarish, while still others are very elongate and narrow. They do not occur any more frequently in the skulls of adolescents than they do in adults, and they are very frequently absent entirely in the former.

As to why these adventitious ossifications occur in one skull and not in another, I have, up to the present time, been unable to discover. They occur with equal frequency in small, contracted skulls, as in unusually large crania, or in the skulls of hydrocephalics, where their presence would seem to be more in demand to insure ossific filling in of the lateral fontanelles.

It is still more puzzling to find a reason for from one to four epipterics occurring on one side and none on the opposite side of any particular cranium in man. We see a similar difficulty for solution to account for no Wormian bones in the lambdoid suture of one human skull, and upwards of an hundred in another, with no apparent reason for their being there.

When published, my paper will take up all these questions more fully, illustrating my researches by various sketches presenting unusual conditions as to the epipterics and the sutures at the lateral aspect of the skull in the Primate.

49. An experimental study of the origin of blood and vascular endothelium in the Teleost embryo. Charles R. Stockard, Cornell University Medical College, New York City.

The study of the origin and development of the blood and vascular endothelium has proven a difficult problem, mainly on account of the fact that the circulating fluids of the embryo usually begin to flow before the cellular elements of the blood are completely formed. These early developing cells are thus quickly washed from their places of origin and diffusely scattered throughout the embryonic tissues. All types of corpuscles are therefore found in intimate association, whether their origins may have been from a common center or from distinctly separate sources. The study of no other tissue presents this obstacle. It has thus seemed highly advantageous to obtain material in which the circulation of the body fluids might be prevented without seriously altering the normal processes of development.

Several years ago I observed that the eggs of the fish, Fundulus, when treated with weak alcohol, chloroform, ether and other solutions developed embryos in which the blood failed to circulate although the heart pulsated in a feeble manner. During the last three years a systematic study of the origin and development of the blood and vessels in embryos with a heart beat but without a circulation has been conducted. This investigation of the experimental material has at all times been controlled by a study of the blood in the normal embryos.

Such material permits the analysis of the following propositions: Do blood corpuscles and vascular endothelium have a common origin from definite anlagen, or does the one arise from a localized anlage and the other from widely distributed sources? Does vascular endothelium ever give rise to any type of blood corpuscles? Do all types of blood corpuscles arise from a common anlage? Do certain organs such as the liver have a true hematopoietic function or simply serve as a seat for the multiplication of blood cells derived from other sources? What rôle does circulation and function play in the normal development and history of blood corpuscles? Finally, the specific question, is the bony fish an exception to the rule that all eggs with meroblastic cleavage develop blood islands in the yolk-sac? All recent workers claim that there are no blood islands in the Teleost yolk-sac.

In many instances the embryo develops in a fashion closely approximating the normal when the heart beat is fairly strong. The blood fails to circulate, however, on account of the fact that the heart is either blind at one or both ends or fails to connect with the veins. In a normal embryo the plasma begins to flow from the vessels of the embryo out into the sinusoids of the yolk-sac, and there establishes a complex vitelline circulation. In individuals in which there is no circulation the plasma accumulates in the pericardial sinus and also in Kupffer's vesicle at the posterior end of the body.

The distended pericardial vesicle forces the head end away from the yolk sphere and thus stretches the heart out into a long straight thread-like structure. These hearts present a great variety of forms depending upon the extent of the pressure in the pericardium. When studied in sections such hearts are in some cases actually solid strings of cells, an inner endothelial string surrounded by a single layer of myocardial cells. In other cases they are slender endothelial lined tubes, while in still others the endothelial cavity is distended and filled with plasma although both ends are closed so that in life the plasma is churned up and down by the pulsation of the heart yet is prevented from escaping or flowing out through the aorta.

The dorsal aortae are in certain specimens almost impossible to identify with high power since their lumina are obliterated—while in other specimens the aortae are well formed endothelial tubes. Yet invariably the aortae never contain any trace of blood corpuscles. The yolk vessels and cardinal veins of such embryos are also distinctly lined with vascular endothelium. It must be concluded from abun-

dant observations that the endothelial vessel linings are present and may arise in all parts of the embryo and do not arise from a local anlage situated in some limited part of the embryo or yolk-sac.

The blood of the Teleost has been found to arise from the so-called 'intermediate cell mass'—Swaen and Brachet, Ziegler, Sobotta and others. These authors differ, however, as to the origin of the cells of the 'intermediate cell mass,' claiming them to be separated from the myotomes, schlerotomes or to be an accumulation of mesenchymal cells. This cell mass in the material here studied is always found to be distinctly connected with and derived from the mesenchyme. Early workers claimed that the blood of the Teleost arose in blood islands on the yolk-sac—but more recent investigators have held that the Teleost forms the marked exception to the rule that in all meroblastic types of eggs the blood arises in islands on the yolk-sac. In the Teleost they hold that the entire blood anlage is within the 'intermediate cell mass.' In Fundulus, however, it is found that blood islands do exist in the yolk-sac and continue their development in this position to give rise to well differentiated masses of erythrocytes when there is no circulation. Normal embryos show these islands distinctly in life and when the circulation is established the corpuscles are swept away in the same fashion as those arising from the intermediate cell mass. The erythrocytes in Fundulus embryos have, therefore, two distinct and limited places of origin, first, in the stem vein or conjoined cardinal veins, and, second, from the blood islands of the yolk-sac.

The stem vein may be single or double, separate cardinals. The blood forming portion is posterior, behind the anterior portion of the kidney and extending into the tail. The yolk islands are always on the posterior and ventral yolk surface and do not extend over the anterior surface.

It is clearly shown by these embryos that the vascular endothelium is of almost universal distribution arising from the mesenchyme, while the blood corpuscles arise from a limited area. The vascular endothelium never gives rise to blood cells. So that the heart, aorta and vessels of the anterior end of the body, although invariably lined with endothelium do not contain a single corpuscle in embryos of any age. Embryos have been studied up to 20 days old; (the normal embryo hatches and is free swimmimg after the 12th day).

The erythroblasts developing from the 'intermediate cell mass' and from the splanchnic layer of mesenchyme in the yolk-sac are not at first surrounded by endothelium. As development proceeds the cells surrounding the mass which are of the ordinary embryonic mesenchymal type differentiate or flatten out to form an endothelial layer surrounding the blood cell mass.

All of the corpuscles arising in the stem vein and yolk-sac develop into erythrocytes. Numerous cells closely resembling poly-morphonuclear and polynuclear leucocytes are found to arise chiefly in the head region and later such cells are found throughout the body. These cells are often very degenerate in appearance and it cannot be definitely

stated what their nature actually is. They occur in the tissue and not within the vessels and are abundant in normal embryos as well as those without a circulation.

The further development of the erythroblasts is significant in embryos without a circulation. These cells reproduce rapidly by mitosis and finally give rise to well formed erythrocytes, the cytoplasm of which contains a normal amount of haemoglobin. The haemoglobin forms in the cells within the stem vein and in the blood islands and attains a bright red color in life. As the embryo grows in size the erythrocytes within the stem vein are further removed from the surface supply of oxygen and after about the eighth day of development they begin to degenerate, and in embryos of sixteen days only a few cells of the erythrocyte type are present in the stem vein along with numerous more or less degenerate mesenchymal cells. The blood islands are better supplied with oxygen and the erythrocytes persist and present a bright red color. When compared with the erythrocytes of a normal embryo those of the blood islands in an old noncirculating specimen show an interesting condition, instead of the healthy, slightly granular nucleus of the fish corpuscle the nucleus is more compact and darkly stained resembling in a striking way the reptilian type of corpuscle or 'Sauroid type' of Minot's classification.

Circulation and normal function seem therefore necessary in order to maintain the typical appearance and structure of the red blood cells in these fish, although such cells originally attain a perfectly typical structure without having circulated.

Finally, these embryos in which the blood cells arise in a normal manner, yet are never permitted to circulate, furnish material for answering in a conclusive way the long contested question—whether the so-called hematopoietic organs such as the liver do actually contain cells which give rise to blood cells, or serve merely to harbor multiplying blood cells in their sinusoids. An examination of the liver of a normal Fundulus embryo of seven or eight days shows it to be very vascular and numerous erythroblasts in mitotic division are often seen. The organ presents the usual hematopoietic appearance.

A similar examination of the liver at any stage of development of an embryo in which the blood has not circulated shows a marked contrast to the normal. The organ is perfectly compact scarcely a vessel is to be found with the highest power on thin sections. *Such a liver does not contain a single erythroblast or erythrocyte in any condition.* The liver of the bony fish has no true hematopoietic function.

50. Experiments on the amphibian ear vesicle. G. L. Streeter, Carnegie Institution, Johns Hopkins Medical School, Baltimore, Md.

At the last meeting of the Anatomists the results of a series of experiments were shown in which the ear vesicle in amphibian larvae was transplanted in other specimens and intentionally placed in an abnormal posture. It was shown that there is a subsequent spontaneous

correction of the posture and that the resulting labyrinth has normal relations to its environment.

Since then the attempt has been made to determine the time at which this spontaneous rotation of the ear vesicle occurs and lantern slides will be shown representing this period and showing the histological conditions under which the rotation occurs.

51. Comparative studies of the neck muscles of vertebrates. R. M. Strong, Department of Anatomy, The University of Mississippi.

During the past five years, I have been engaged in comparative anatomy studies which have concerned birds especially, but have included Necturus and the alligator also. Publications are in process of preparation for all of these animals, and a work on the anatomy of the Tubinares (albatrosses, petrels, etc.) is approaching completion.

For this occasion, I have selected one of the sections I found less satisfactorily treated in the literature. I have been especially impressed with the need of work on the neck muscles, and I have been unable to find satisfactory illustrations or descriptions of some of these structures. Even the publications of Owen, Gadow, Fürbringer, and Shufeldt have omitted much, and Fürbringer has considered the neck muscles only incidentally. The present confusion in terminology has impressed on me more than ever before the great need of action by a commission to extend the B N A to comparative anatomy.

As the structures described in this paper can not be satisfactorily described without illustrations, no account of them appears in this abstract. Lantern slides and demonstration.

52. Further observations of the origin of melanin pigments. R. M. Strong, Department of Anatomy, The University of Mississippi.

In two previous publications the writer has presented evidence supporting the position maintained by a number of writers that epidermal pigments are produced *in situ*, i.e., are not of dermal origin. A little over two years ago, I assigned to Miss Katherine Knowlton, a graduate student at The University of Chicago, some work on the pigments of feather germs from Plymouth Rock and Brown Leghorn fowls.

In the course of the studies, some interesting evidence concerning the origin of melanin pigments was obtained. Chromatophores were found in the dermal pulp near its proximal end as well as in their usual location in the epidermal cylinder of the feather germ. We found no evidence, however, that these dermal chromatophores wander into the epidermis. They differ in form and size from the epidermal chromatophores. They were also never seen in positions that would indicate migration into the epidermis, although a few were found crowded against the basement membrane which was not penetrated at any point. These dermal chromatophores occur mostly at a level lower, i.e., earlier in the development of the feather elements, than the chromatophores which supply the melanin pigment located in the feather elements.

It seems probable that these chromatophores are comparable to those of the skin dermis. They were found in a more or less continuous series leading to the inferior umbilicus, and it is probable that this continuity would have been complete with the chromatophores of the skin if the preparations had included the tissue which surrounds the feather follicle. A more complete account of this work will be published later. Demonstration of miscroscope slides with sections of feather germs which show dermal pigment.

53. The date and clinical significance of fusion of the costal element with the transverse process in the seventh cervical vertebra. T. WINGATE TODD, from the Anatomical Department, Western Reserve University, Cleveland, Ohio.

In a paper presented at a meeting of the Anthropological Society of Paris (Bull. et Mém. de la Soc. Anthrop. de Paris, 1914) on the variations of the transverse process of the seventh cervical vertebra in Homo, I discussed at length the evidence afforded by the examination of some three hundred vertebral columns regarding the precise disposition of the costal element and its relation to the foramen transversarium (B N A). While many accounts mention the appearance of an ossific centre in the costal element of the transverse process of this vertebra, they do not agree as to the precise date of its appearance, from which we may reasonably gather that the date varies greatly from individual to individual. Concerning the date of fusion of the costal element with the true transverse process, information seems to be even more scanty. That the matter has some importance, however, is apparent when one considers the confusion existing in the minds of many clinicians and anatomists regarding the mutual relations of the transverse process of the seventh vertebra and its cervical rib or costal element.

So much more frequently are cervical ribs observed in children than in adults that Dr. Gilbert Scott has been led to state his belief, for which he says he has some evidence and is collecting more, that the osseous tissue of the cervical rib is absorbed as the child grows (Brit. Med. Journ., 1912, vol. 2, pp. 483–84). This extraordinary statement led me to investigate carefully the date of fusion of the costal element with the transverse process in the belief that such fusion is the explanation of the so-called absorption. Skeletons of suitable age are, however, not readily obtained, but that fusion may be delayed until comparatively late in adolescence is shown by the presence of an unfused costal element, which by no stretch of the imagination could be called a cervical rib, in an Austrian male of 18 years. Fusion is not always delayed so late as this, for in a negro male 17 years of age I found the fusion already complete. In neither of these skeletons had the epiphysis of the spinous processes yet fused with the main part of the bone. The striking difference between the two skeletons is that while the seventh vertebra in the negro approximates the sixth in type, that of the Austrian more nearly resembles the first thoracic. It may be that such a difference in type is closely allied with the date of fusion.

It is plain, however, that fusion may be delayed until the approach of maturity.

Extending my observations to the Primates, I found further evidence in favor of the foregoing conclusions. In a female gorilla which I estimate to be 14 years old, since the third molars are just being cut, the costal element of the seventh vertebra had fused with the transverse process on the left side, but was still incompletely fused on the right. In a specimen of Ateles belzebuth which I believe to be about five years old, as the permanent dentition is almost completed (i.e., the third molars are emerging from the alveolar process and the canines are not quite fully erupted) while the epiphyses of the limbs are still ununited with the shafts of the bones, the fully ossified costal element of the seventh vertebra is distinct and separate from the true transverse process. Both of these animals were on the verge of maturity.

The examination with the Röntgen rays of fresh skeletons of children, which exhibited distinct costal elements unfused with the transverse process in the seventh vertebra, clearly showed that the skiagram is not to be trusted on the point. Its evidence may be equivocal and too indefinite to decide whether fusion has occurred or not.

Hence one may feel justified in stating that so-called absorption of infantile cervical ribs as the child grows is probably explained by the fusion of the fully ossified costal element with the transverse process, an occurrence which may be delayed till after puberty, and the date of which cannot be indicated with certainty by the aid of the Röntgen rays.

54. Is function and functional stimulus a factor in producing and preserving morphological structures? EDUARD UHLENHUTH.

I propose to show you some preparations of a series of experiments, which I have made with the transplanted eyes of Salamander maculosa. These experiments are designed to show whether or not organs of a typical functional structure, after transplantation to an abnormal site, require function or functional stimulus, in order to be preserved at this new site. In short, I wished to investigate whether function and functional stimulus is a factor in preserving functional structures in transplanted organs.

After cutting through the optic nerve the eyes of amphibians undergo considerable degeneration of the retina. Later, however, the optic fibres regenerate and the trunks of the optic nerve unite. At the same time the retina which had undergone disintegration becomes restored to a normal condition.

By transplanting the eye to a place far removed from the normal position such a reunion of the optic nerve with the central nervous system is inhibited and the eye is permanently prevented from functioning. Partly, as might be supposed, the functional stimulus, namely light, which influences the retina even after transplantation, could bring about this result. I therefore used two series of larvae with transplanted eyes, both consisting of more than 100 specimens. One of the

series was kept in ordinary daylight and the other series in a dark room, and in the latter case, no light, not even red light, was admitted while the animals were being cared for, by which means the retina could neither functionate nor be affected by functional stimulus. The two series were then compared.

I will show you preparations of these two series. Those of the daylight series, as you will see, clearly show a regular increase in degeneration during the time immediately following upon transplantation, succeeded by a second period of a gradual increase in regeneration. To each preparation of the light series belongs one preparation of a transplanted eye of the same age but kept in darkness, and this is, as you will find, always much more normal than that of the light series. If function or functional stimulus were to favor regeneration the relation would be the reverse.

But I might have selected for each light eye a less normal dark eye of the same age, instead of a more normal one. As in both series the same factor, but in a contrary sense, has equally influenced every transplanted eye, these variations of the eyes of the same age and series cannot be explained as an effect of this factor, but must be the result of another factor which is variable and not controlled and which is perhaps produced by slight variations of the position of the eyes in the new place. Anyway, by having a large number of experiments, one is able to take the average of every age of both series, by means of which I obtained curves of the rapidity of regeneration in every series. Compared with the other they do not show any differences which can be ascribed to the influence or absence of light.

If the function or functional stimulus does nor influence the rapidity of regeneration it might be impossible to keep the transplanted eyes permanently in a normal condition without these factors; but you will find fairly old transplanted eyes, if you will look at the preparations. Some of them were made after the eye had been left for fifteen months in its new position. You will see that even those old eyes were perfectly normal.

The conclusions, therefore, which we must raise from these facts is the following:

The regeneration of the structures of vision following destruction caused by the transplantation of the eyes can by no means be considered functional regeneration, and even the rapidity of this regeneration is not influenced by functional stimulus. The permanent preservation of the retina, when deprived of all function or functional stimulus and kept in perfectly abnormal conditions, is not a process which is governed by functional adaptation.

55. On the early development of the inguinal region in Mammalia. JOHN WARREN, Harvard Medical School, Boston.

The early development of the gubernaculum and processus vaginalis was studied in the human embryos and in the embryos of the cat, sheep, rabbit, and rat in the Harvard Embryological Collection. As far as

possible different stages in each form were described so as to show the more important changes in the early development of this region.

In human embryos the first sign of a connection between the Wolffian body and the abdominal wall can be seen in an embryo of 13.6 mm., transverse series no. 859. From the cells in the wall of the Wolffian duct a fairly distinct mass of cells may be traced dorso-laterally immediately beneath the primitive peritoneum covering the Wolffian body to join the abdominal wall at a slight elevation, the inguinal crest. This cell mass invades later the abdominal wall, and is the anlage of the gubernaculum. A fossa is formed in the peritoneal cavity dorsal to this primitive gubernaculum. In an embryo of 16.6 mm., transverse series no.1707, the cells in the gubernaculum have become much concentrated, and have begun to invade the ventro-lateral abdominal wall, where as yet there is no differentiation of the abdominal musculature. The gubernaculum steadily increases in density and length, and in an embryo of 19. mm., transverse series 819, it extends well through the abdominal wall, where its peripheral end expands and blends with the cellular constituents of the wall. At this stage the musculature ventrally forms a general cell mass, but more dorsally has differentiated into its three layers. In embryos of 22.8 mm., frontal series no. 757, and 24 mm., transverse series no. 38, the musculature is completely formed and grows around the peripheral part of the gubernaculum. An indistinct prolongation of the latter can be traced through the external abdominal ring into the subcutaneous tissue of the abdominal wall, forming a primitive ligamentum scroti. The processus vaginalis appears soon after this stage in embryos of 37 mm., transverse series no. 820, and 42 mm., transverse series no. 838, and develops downward and inward chiefly on the mesial side of the subernaculum.

In the Carnivora, the primary point of contact between the Wolffian body and the abdominal wall can be distinguished in a cat embryo of 12 mm., transverse series no. 399. Here the anlage of the gubernaculum resembles the earliest form in human embryos, and a shallow fossa is formed also on its dorsal side. In an embryo of 15 mm., transverse series no. 436, the abdominal portion of the gubernaculum has increased in length, while the inguinal or muscular part has already passed through the abdominal musculature into the subcutaneous tissue over the external ring. The musculature is fairly well differentiated, and the external ring can be clearly seen with the peripheral end of the gubernaculum projecting through it. An embryo of 31 mm., transverse series no. 500, gives an excellent view of the whole length of the gubernaculum. The abdominal portion is now very long and slender, and the scrotal part appears as a large oval mass projecting through the external ring. The processus vaginalis has just begun to develop, but is growing downward on the lateral side of the gubernaculum. In an embryo of 39 mm., the processus vaginalis surrounds the gubernaculum except on its dorsal side, and can be traced well into the large expanded scrotal portion beyond the external ring. This represents the most advanced stage of any of the embryos studied.

The sheep embryos offered the best view of the development of the region in ungulates. The earliest appearance of the gubernaculum was seen in an embryo of 18 mm., transverse series no. 1238. It forms a thick connecting bar of tissue, similar to the corresponding stage in the cat and in man. Its extension through the abdominal wall, the development of the abdominal musculature about it, and the earliest trace of the processus vaginalis follow closely the course already described in the cat. The processus vaginalis develops first on the lateral aspect of the gubernaculum and then seems to grow downward chiefly in its substance. This is rather strikingly shown in the oldest embryo, transverse series no. 1696, 48.4 mm., where the lower end of the processus vaginalis is completely embedded in the large expanded distal end of the gubernaculum, into and about which the cremaster fibers can be seen growing. The relation between the processus and the gubernaculum is different at these stages from any of the other forms.

In rodents, a rabbit embryo of 11 mm., transverse series no. 1327, shows a primary point of contact between the abdominal wall and the Wolffian duct exactly similar to the cat embryo of 12 mm. A rat embryo, transverse series no. 1823, 9.3 mm., shows the early gubernaculum at a stage a little further advanced and comparable to that of the human embryo of 16.6 mm., and to the sheep of 18 mm. In all cases the presence of the retro-gubernacular fossa in the peritoneal cavity is striking. The development of the gubernaculum and processus vaginalis is very precocious in rat embryos and seems to advance more rapidly in the earlier stages than in those of the other mammalian embryos studied. A rabbit embryo of 21 mm., transverse series no. 738, gives an excellent view of the whole extent of the gubernaculi of both sides and the three parts—abdominal, inguinal or muscular, and scrotal—are clearly differentiated. There is no sign yet of the processus vaginalis which can be seen however in a rat embryo of 15.2 mm., transverse series no. 1801, appearing on the lateral aspect of the gubernaculum. The processus vaginalis begins to develop in rabbit embryos of between 21 and 29 mm., and in the latter arises from the large funnel-shaped fossa dorsal to the gubernaculum and extends downward on the dorso-lateral side of the gubernaculum. This ends in a dense but narrow ligamentum scroti that fuses in the subcutaneous tissue with the one of the opposite side. The cremaster fibers are well marked and arch over and blend with the gubernaculum as in the older sheep embryos.

56. Is defective and monstrous development due to parental metabolic toxaemia? E. I. Werber, Department of Biology, Princeton University.

The problem of the causes underlying defective development has recently received masterly treatment by F. P. Mall. In his extensive study based on 163 pathological ova he supports the conclusion arrived at by the experimental embryologists, namely, that the human monsters are—with the exception of the hereditary 'merosomatous' terata—due to injurious influences of atypical environmental factors. He

makes the specific suggestion—which seems justified in the light of evidence brought forth by him as well as by clinical data—that the monstrous development of some ova may be due to their inadequate nuitrition owing to the imperfect implantation in a diseased uterus. It would seem that this hypothesis will hold good at least for many pathologic embryos aborted during the first two months of pregnancy. At any rate, the principle advocated by the hypothesis, viz., the influence of unusual environmental factors, seems to be correct beyond any doubt.

Mall's interpretation could not, however, be applied to monstrous fetusses of the later months of pregnancy or to monsters after full-term births. Some environmental factors must be looked for other than faulty implantation of the ovum, to account for the occurrence of such cases. The results of investigations in experimental embryology and teratology by Dareste, Roux, Hertwig, Féré, Morgan, Tur, Stockard, and others, who obtained monstrous development of ova which had been subjected to the action of physical and chemical modifications of the environment, suggested to me that the human as well as other mammalian monsters may be due to the physical or chemical action of some substances in the blood of one of the parents on either one of the germ cells or on the fertilized ovum respectively. The toxic substances found in the blood of invididuals afflicted with some diseases of metabolism, I think, might be the ones which could be made responsible for the origin of monstrous development.

To test this hypothesis it would be necessary to breed mammals in which certain diseases of metabolism had been produced experimentally, for the spontaneous occurrence of these disturbances in animals is too rare to permit of conclusive breeding experiments. On the other hand, to produce these diseases experimentally, at least as far as this can be done by the present rather inadequate methods of experimental pathology of metabolism, requires some facilities which, so far, are beyond my reach. I have thus had to confine myself to a preliminary step in the investigation.

This consisted in subjecting the eggs of an oviparous vertebrate to the action of solutions of substances found in the circulation of man under certain pathological conditions of metabolism. The eggs of a Fundulus heteroclitus were chosen as the object of experimentation and they were subjected in early (1 to 2 or 2 to 4 cells) cleavage stages to the action of sea-water solutions of various strengths of urea, butyric acid, lactic acid, acetone, sodium glycocholate and ammonium hydroxide, respectively. Positive results permitting of definite conclusions were so far obtained only with butyric acid and acetone.

For butyric acid 10 cc. of a $\frac{1}{12}$ to $\frac{1}{14}$ gram molecular solution, added to 50 cc. of sea water was found to give the best results, that is, the greatest number of monsters, while very much stronger solutions of acetone, namely, about 35 to 40 cc. in 50 cc. of sea-water were found to be necessary to obtain like numbers of monsters. In the case of butyric acid a long exposure was found to kill most eggs and it was necessary to limit it to 20 hours, while in the case of acetone the eggs could be kept

in the solution up to 48 hours, exposures longer than this increasing their mortality. It was also found that the eggs were more susceptible to the influence of these chemical modifications of the environment during the first and second than during the third and fourth cleavages.

The results obtained with butyric acid and acetone solutions are very much alike. Great numbers of cyclopean monsters were found in both these series of experiments. I have recorded in my observations the occurrence of transition from two normal eyes in the typical position in the head all the way down through the more or less closely approximated eyes or eyes of a double composition and through true cyclopia to complete anophthalmia as described by Stockard in his experiments with magnesium chloride and alcohol. Stockard has also described a peculiar change in the form and position of the mouth in the cyclopean embryo. The mouth in such embryos has the appearance of a snout, a proboscis-like structure and is pushed down below the cyclopean eye. This displacement into the ventro-lateral position Stockard attributes to the circumstance that the cyclopean eye, being frontally located, has caused the mouth to move downward. I have observed this occurrence in most of the cyclopean monsters found in my experiments as well as in many cases of dorsal microphthalmia or even in some cases of asymmetric monophthalmia.

Besides the cyclopean, asymmetrically monophthalmic, microphthalmic and anophthalmic embryos there was found a very great variety of monstrosities in which practically the entire bodies of the embryo were more or less involved in the malformations. Thus curiously misshapen dwarfs with vestigial eyes or blind or very elongate, greatly malformed embryos, often with many waistlike constrictions were of not infrequent occurrence. Eggs in which only an anterior part of the embryo ('meroplastic embryos'—Roux), as if the rest of the body had been mechanically removed, were found in great numbers. Of these the ones in which an approximately anterior one-half of the body was present would correspond to the 'hemiembryones anteriores,' which Roux obtained experimentally in the frog by injuring one of the blastomeres after the first cleavage. These hemiembryos found in my own experiments have defective or sometimes very rudimentary eyes, they often exhibit evidences of oedema and are as a rule extremely misshapen. Eggs were also recorded in considerable numbers in which only a malformed head or small anterior part of it could be observed. These 'meroplasts' are so deformed that only by the presence of rudimentary eyes is it possible to determine that they are heads.

But the most curious and most significant of all meroplasts recorded were eggs in which all that could be observed on the yolk-sac was a very small fragment of brain tissue with a *solitary eye*, which was sometimes somewhat defective—of the "coloboma" type. The fragment of brain tissue is usually smaller than the eye it has given rise to. Since the eggs were fixed by Child's sublimate-acetic method and preserved in formaline, the embryos are white while the yolk-sacs are transparent; yet nothing at all can be seen in the transparent yolk-sac to indicate that the embryo had sunken into it leaving one of its

eyes on the surface where it might have been constricted off. Moreover, in order to establish the fact of the occurence of the solitary eye beyond any possibility of skeptical criticism, I have sectioned one of these eggs on which besides the solitary eye several very small fragments of tissue could be observed in the living as well, as in the fixed specimen in various places, distant from each other, on the yolk-sac. The interpretation proved to be perfectly correct. *For*, besides the referred to few, small amorphous fragments of tissue scattered over the yolk-sac *there can be seen only an eye typical in all its structures*, while nothing can be found, to indicate the presence of an embryo. *This is, as far as I am aware, the first case on record of the independent development* ('self-differentiation'—Roux) *of the eye.*

Another instance of apparently independent development of the eye in these experiments has occurred in some cases where an eye appeared at a considerable distance from a monophthalmic embryo.

The ear vesicles are often involved in the malformations of embryos. This can be readily seen by their sometimes enormous size. Some asymmetrically monophthalmic embryos after hatching could not swim directly forward, dropping to the bottom of the dish in which they were kept, if forced to do so. They could only move in circular or spiral lines which would indicate some injury to the semi-circular canals.

There is a wide range of variation in the deformities of the blood vascular system. The heart is almost perfect in some embryos in which cyclopia is the only superficially noticeable defect. In cases of more extreme malformation the heart may be only a very delicate, straight tube in some embryos while it may be absent altogether in others. In this connection it may be of interest to note that I have found some eggs in which all that was present of the embryo was a functioning heart and some rudimentary blood vessels. The degree of malformation of the blood vessels is subject to a great deal of variation. There may be merely blood-islands scattered on the yolk-sac, rudimentary, imperfectly connected, or in some instances more or less normal vessels.

The tendency of butyric acid and acetone solutions to produce twins seems to be only slight for I have observed only a few of such cases. I have only one case comparable to the "Siamese twins" type of the human. In this egg two deformed embryos with a common heart had developed on opposite sides of the egg. Other cases of twin formation found in these experiments belong to the type known as 'duplicitas anterior,' which was produced experimentally by mechanical means by Speemann.

Not infrequent is the occurrence of amorphous embryos or small amorphous fragments of tissue on the yolk-sac as the only evidence of development.

The mechanism involved in the action of butyric acid and acetone in bringing about these effects will have to be taken up as one of the further steps of this investigation. At the present time I can only state that there seems to occur in the eggs when subjected to the action of butyric acid and acetone solutions an elimination of substance of the blastomeres or possibly of the germ-disc. This elimination may be brought about either by precipitation or by the solvent effect respectively of the chemicals used in the experiments. Whatever parts of the

blastoderm survive that process of destructive elimination, may go on developing to form an isolated organ or a part of the body or a complete embryo with defects in some organs.

57. The ileo-jejunal artery. C. R. Bardeen, University of Wisconsin.

From the superior mesenteric artery opposite the origin of the ileocoecal artery there arises a branch which passes to the free small intestine near the junction of the upper with the middle third of the gut as measured from the duodenum to the coecum. This branch may be called the ileo-jejunal artery. In a previous article (C. R. Bardeen, "The critical period in the development of the intestines," American Journal of Anatomy, vol. 16, p. 427) I have shown the probability that the region supplied by this artery represents the junction between that portion of the small intestines the primitive coils of which develop within the umbilical cord, the ileum, and that portion the primitive coils of which develop within the abdominal cavity, the jejunum. A study of the blood supply of the intestines in a number of fetuses and adults has shown that the portion of the intestines proximal to the center of the region supplied by the ileo-jejunal artery, the 'jejunum,' varies from 26 to 44.1 per cent of the total length of the small intestines as measured on the side opposite the mesentery and from 27.4 to 41.2 per cent measured on the mesenteric border.

In 10 fetuses measuring from 40 to 280 mm. in length (vertex breach) the average proportional length for the jejunum opposite the mesenteric border was 37.1 per cent with extremes of 32.2 and 44.1 per cent. In only two cases was it 40 per cent or over.

In 18 adults varying in age from 14 to 74 years the average proportional length of the jejunum was 33.5 per cent with extremes of from 26 to 40.3 per cent. In five cases it was under 30.7 per cent and in two cases 40 per cent or over. In eleven instances the jejunum as measured in the mesenteric border averaged 33.3 per cent with extremes of from 27.4 per cent to 41.2 per cent. In four instances it was less than 30 per cent; in two instances 40 per cent or more.

In the adult specimens examined the length of the intestines varied from 138 to 294.5 inches but I have been able to trace no correlation between the length of the free intestines and the proportion between the proximal and distant portions. Aside from the average greater proportional length of the jejunum in the fetuses examined as compared with the adult I have found no correlation between age and the proportional length of the jejunum and with a greater number of specimens examined this difference might disappear. In 8 women the average length of the gut was shorter than that in 10 men, although I have found no certain correlation between length of gut and length of body. In the 8 women the average length of the jejunum was 31.96 per cent (extremes 26, 40 per cent) and in the 10 men, 35.31 per cent (extremes 27.3, 40.3 per cent). This may indicate a tendency on the part of women to have a longer ileum than men have and may possibly be related to the greater tendency in women to constipation.

THE FOLLOWING DEMONSTRATIONS WERE SHOWN:

1. *Certain aspects of hematogenesis in the pig embryo.* V. E. EMMEL, Department of Anatomy, Washington University Medical School.

The microscopical demonstrations and drawings are to illustrate data relating: (a) to the cytological structure and morphological relations of the cell clusters in the dorsal aorta (cf. also abstract of paper on the same subject) and the macrophags and mesamoeboids in the liver sinusoids and coelomic cavities, with reference to the problem of the relation of endothelial tissue to hematogenesis in these regions, and (b) to certain cytological characteristics of erythrocytes during their cytomorphosis.

2. *Sections, models and drawings showing the development of the chondrocranium in Felis domestica.* R. J. TERRY, Washington University Medical School.

The following points of interest have been selected for demonstration:

(1) Basal plate: the position of the notochord, flexures of the basal plate.

(2) Occipital region: basal and lateral moieties of occipital condyle, hypoglossal canal, dorsal root and ganglion of the hypoglossal nerve, the occipital hypochordal arch, neural arches of the atlas and atlantal foramen.

(3) Otic region: evidence of independence of chondrification of otic capsule, relations of suprafacial commissure, independent chondrification of the parietal plate, development of the tegmen tympani, course of the facial nerve, formation of the internal acustic meatus, foramen cochleae and aquaductus cochleae, cavum supracochleare, supraganglionic cartilage, distribution of the acustic nerve.

(4) Orbito-temporal region; hypophyseal cartilage, development of the dorsum sellae, foramen hypophyseos, early relations of the ala orbitalis, primary elements of the ala temporalis, origin of the foramen lacerum, epipteric cave, relations of ocular muscles to chondrocranium.

3. *Microscope slides showing feather germs with dermal pigment.* R. M. STRONG, The University of Mississippi, Oxford.

4. *Photographs of plates illustrating the anatomy of the albatross (Diomedea).* R. M. STRONG, The University of Mississippi, Oxford.

The original drawings were made by Mr. Kenji Toda, artist for the Department of Zoology at the University of Chicago. About one-third of the plates are represented in the exhibit.

5. *Photographs, drawings and charts illustrating (A), the morphology of the mammalian seminiferous tubule and (B), the relation of the stages of spermatogenesis to the tubule.* GEORGE M. CURTIS, Vanderbilt University Medical School, Nashville.

6. Demonstrations of 'endothelioid' cells. Hal Downey, University of Minnesota, Minneapolis.

1. Normal lymph node of guinea-pig; Helly, methyl green and pyronin. The sinuses contain many of the so-called 'endothelioid'. cells, both attached and free.

2. "Endothelioid" cells in lymph node from normal cat; Helly, May-Giemsa. They are large protoplasmic cells which form a part of the reticulum. The wall of the small lymph sinus is in part composed of processes from these cells. One of the cells has almost completely separated from the reticulum. Its nucleus is indented and it has phagocytosed a red corpuscle. This method does not bring out the reticular fibers. The reticulum is not covered by an endothelium; if it were it should be possible to see the nuclei of its cells. The large cells will separate from the rest of the reticulum and form the 'endothelioid' cells of pathologists.

3. Lymph node from normal cat, stained by one of the methods for reticular fibers. The 'endothelioid' cells contain fibers in their peripheral portion.

4. Spleen from Mandelbaum's case of Gaucher's disease; Orth's fluid, iron-hematoxylin, fuchsin S, orange G, toluidin blue. 'Endothelioid' cells very numerous in the pulp and venous sinuses.

5. Lymph node from Mandelbaum's case of Gaucher's disease; alcohol fixation, stain as for (4) above. This field shows that the large, characteristic 'endothelioid' cells are derived from the reticulum and not from endothelium. The long strand in the center of the field is a part of the modified reticulum, and the long strand of reticulum approaching the center from the right gradually assumes the character of the protoplasm of the Gaucher cells.

6. Lymph node from a case of Hodgkin's disease; Helly, Weigert's iron-hematoxylin, fuchsin S, orange G, toluidin blue. In many cases the fibers of the reticulum can be seen to penetrate the 'endothelioid' cells.

7. A differential counterstain for vertebrate embryos. W. A. Willard, University of Nebraska, College of Medicine.

Pig embryos designed chiefly for class study of organogenesis are first deeply stained *in toto* with borax carmine then cut into serial sections 20 micra or more in thickness and counterstained on the slide with a dilute solution of Lyons blue in absolute alcohol which has been rendered a blue-green color by the addition of a few picric acid crystals The strength of the solution and the time required to stain is best determined by experiment with any particular lot of material. The result of the counterstain is to add brilliancy and transparency to the whole section slightly decolorizing the carmine and giving a selective stain of light green to the developing nerves and certain portions of the central nervous system Blood cells and to a certain degree epithelial structures are differentiated. By this method short series are made available with a minimum amount of handling, recommending it as a practical laboratory method.

8. A double embryo of the spiny dogfish (Squalus acanthias). W. A. Willard, University of Nebraska, College of Medicine.

This is an example of two normally developed embryos attached by separate yolk stalks to a common yolk-sac. The embryos are in the second year of their intra-uterine development, in what is known as the 'pup' stage, one measuring 16 cm., the other 14.5 cm. in length. The larger of the two is supplied from a larger yolk-sac area as indicated by the vascularization by the vitelline vessels. An exposure of the viscera does not disclose any modification of the normal arrangement of organs, such as transposition or reversal of the normal symmetry. As the embryos had slipped from the cloaca of the female before they were noticed no data were obtainable as to position in the uterus or as to other embryos of the same brood.

9. Slides of yolk-sac of 10 mm. pig embryo. H. E. Jordan, University of Virginia, University.

Technic (1): Zenker fixation, hematoxylin-eosin stain; slides of yolk-sac of 4 mm. pig embryo. Technic (2): Helly fixation, Giemsa stain.

10. Injections of the lymphatics of the lung. Robert S. Cunningham, Johns Hopkins Medical School, Baltimore.

11. Injections of the vascular system in early pig and chick embryos. Florence R. Sabin, Johns Hopkins Medical School, Baltimore.

12. Cell groups of the hypothalamus in man. Edward F. Malone, University of Cincinnati, Cincinnati.

(1) Nuclei tuberis laterales; (2) Ganglion opticum basale; (3) Nucleus paraventricularis hypothalami; (4) The three cell groups of the corpus mammillare.

13. Microscopic preparations showing the reactions of transplanted eyes in Amphibia. Edward Uhlenhuth, Rockefeller Institute, New York City.

14. A human embryo of 22 somites (models and figures). Franklin P. Johnson, University of Missouri, Columbia, Mo.

15. Models of the liver veins of pig embryos. Franklin P. Johnson, and T. F. Wheeldon, University of Missouri, Columbia, Mo.

16. Models of the heart of a 20 mm. pig. T. F. Wheeldon, University of Missouri, Columbia, No.

17. Dissections showing origin, course and distribution of nervus terminalis in the human fetus. Rollo E. McCotter, University of Michigan, Ann Arbor, Mich.

18. Models of the early development of the inguinal region and of the pelvic outlet in human embryos. JOHN WARREN, Harvard Medical School, Boston.

19. Demonstration of reconstructions of lateral hearts and foregut in Citellus to show connection of endocardium to entoderm. THOMAS G. LEE, Institute of Anatomy, University of Minnesota.

20. Models showing the development of the hypophysis in Squalus acanthias. E. A. BAUMGARTNER, Washington University Medical School, St. Louis.

The study of the hypophysis, begun at the University of Minnesota and completed at Washington University Medical School, is represented in part by a series of ten models.

A model of a 19 mm. embryo shows Rathke's pouch extending obliquely forward and dorsalward from the mouth. In a 21 mm. embryo a part of the wall of the oral cavity ventral to Rathke's pouch has begun to evaginate to form the anterior end of the hypophysis. From these two out-pouchings are formed the hypophysis of the adult. The upper lateral portions of Rathke's pouch are somewhat dilated. In a model of the hypophysis of a 22 mm. embryo, the anterior end is distinctly evaginated; while in a model of the hypophysis from a 28 mm. embryo this part is constricted off from the mouth except for a small stalk connected to its caudal side. Most of the first out-pouching, or Rathke's pouch, will form the caudal end of the anterior lobe. The lateral dilated portions are separated from the median part by two furrows which have appeared on the anterior side of the upper part of the hypophysis. The extreme tip of Rathke's pouch is somewhat enlarged, the anlage of the superior lobe of the hypophysis. A model of the hypophysis of a 33 mm. embryo shows a short anterior end connected by a narrow mid-part to the wider caudal end. A slight ridge, superior to the hypophyseal stalk, connects the lateral portions which, in the adult, form the inferior lobes. In a 48 mm. embryo the model shows that the hypophyseal stalk has disappeared. The superior lobe has two lateral wings extending forward and slightly dorsally. In a model of the hypophysis of a 95 mm. embryo the anterior lobe is very long and the wider anterior and caudal extremities are marked. The inferior lobes project laterally and from their median connection a slender duct connects them to the caudal extremity of the anterior lobe. In the pup stage ridges indicate the beginning glandular structure. These are present on the ventral wall of the anterior extremity of the anterior lobe and on the roof of the inferior lobe. A model of some of the glands of the anterior lobe shows them connected to the ventral wall. They are short-branched tubular outgrowths showing anastomoses. The lumina may not be continuous throughout the anastomosed tubules.

21. Wax models in verification of the nucleus-plasma relation of nerve cells. David H. Dolley, University of Missouri (introduced by E. R. Clark).

As a result of the averages of measurements, for the most part on the crayfish and the dog, the nucleus-plasma norm of functionally resting nerve cells of the same type has been found to be represented by a close numerical constant in all individuals within any particular species.

For final verification, calculations were made for individual Purkinje cells of several dogs from serial sections at 2 and 1μ. From these serials wax models were reconstructed and the data were afforded for the mathematical application of the prismoid formulas. In the case of the wax models, the proportion by weight of wax nucleus to wax plasma is identical within very narrow limits, whatever the size or shape of the cell or whatever the size of the animal or its age between the full development of the relation and senescence. The uniformity of the results after all three methods, with the support of certain collateral evidence, has led to the induction of a law of species identity of the nucleus-plasma norm for corresponding nerve cell bodies (Jour. Comp. Neur., vol. 24, October, 1914).

The shifts in absolute and relative size in nucleus and plasma which result from function, as determined by average measurements, are also confirmed by the application of the wax reconstruction and the prismoid formulas to the individual cell.

22. On the use of orcein as a bulk stain for elastic fibers. A. G. Pohlman,* University of St. Louis.

Blocks of tissue are run through to 95 per cent alcohol and placed in Unna's orcein solution for 2 to 12 hours according to size. Remove to absolute alcohol slightly acidulated with HCl for 2 to 3 hours and then into an excess of absolute alcohol for 6 to 12 hours. The tissue may now be handled in the usual way for paraffin and celloidin technique. If tissues are not sufficiently differentiated after sectioning use 5 per cent oxalic acid. The stain is very resistant and will not be affected by ordinary laboratory methods.

Demonstration I. (1) Plain bulk orcein; (2) Paracarmine followed by orcein; (3) Orcein followed by hematoxylin-eosin; (4) Orcein followed by hematoxylin-eosin and orange G.; (5) Orcein followed by hematoxylin and picrofuchsin; (6) Orcein followed by picrofuchsin; (7) Differentiation in cleared bulk specimen; (8) Differentiation shown in uncleared bulk specimen.

Demonstration II. (1) Length section of Platner's ligament; (2) Length section through drum ligament in chick; (3) Section through attachment of Stapedial plate and membrane of the Fenestra cochleae; (4) Dissection of the gross relations of the columella in chicken, duck, goose and turkey. Platner's ligament shows as a delicate fiber running forward from columella to quadrate bone.

23. Plaster casts of the sphenoid, maxillary and frontal sinuses, the cubical capacity and superficial area of these sinuses. HANAU W. LOEB, St. Louis University.

The casts are made by joining together plaster moulds of those portions of the sinuses lying adjacent to one another in serial sections of the head. The casts are then boiled in paraffin and the cubical capacity determined by ascertaining the amount of water displaced by them. To determine the superficial area, the casts are covered with strips from a known amount of adhesive plaster. The difference between the known amount and that remaining gives the superficial, subject to whatever error results from lack of complete approximation of the strips. This is exceedingly small indeed.

24. A method for handling paraffin sections. IRVING HARDESTY, Tulane University.

A method for staining and issuing paraffin sections for mounting by the students in large classes in histology. Thin sheets of a modified form of celluloid may be obtained under the commercial name, "la cellophane." These sheets are quite thin, perfectly transparent and resist the action of water, alcohol of all strengths, ether, chloroform, and all the oils commonly used in clearing and differentiating, including creosote and oil of cloves. La cellophane of thickness No. 253 may be obtained in sheets $17\frac{1}{2}$ by 25 inches. These may be cut into sheets of desired size. The paraffin sections of a specimen, in sufficient number to supply the class may be placed upon the sheets, straightened out and fixed by the usual albumen-water method. After drying, the entire sheet is treated for staining, clearing and mounting, just as is a slide with a single section. After clearing, the sheet is clipped into small pieces each bearing a section and these pieces issued to the class La cellophane No. 253 is sufficiently thin for the purpose; No. 252 however, is said to be of thinner weight. For work with oil immersion objectives under cover—glasses of ordinary thickness, the student should be advised to mount the pieces with the sections uppermost. The sheets retain practically none or very little of the stain after hematoxylin and anilin dyes commonly employed.

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Allen, William F., A.M., Instructor of Histology and Embryology, *Institute of Anatomy, University of Minnesota, Minneapolis, Minn.*

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Badertscher, Jacob A., Ph.M., Ph.D., Instructor in Anatomy, Indiana University School of Medicine, *517 N. Lincoln Street, Bloomington, Ind.*

Bartelmez, George W., Ph.D., Instructor in Anatomy, *Chicago University, Chicago, Ill.*

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Baumgartner, Edwin A., A.M., Instructor in Anatomy, *Washington University Medical School, St. Louis, Mo.*

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Bayon, Henry, B.A., M.D., Associate Professor of Anatomy, Tulane University, *2212 Napoleon Avenue, New Orleans, La.*

Bean, Robert Bennett, B.S., M.D., Professor of Gross Anatomy, *Tulane University of Louisiana, Station 20, New Orleans, La.*

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Bevan, Arthur Dean, M.D. (Ex. Com. '96–'98), Professor of Surgery, University of Chicago, *2917 Michigan Avenue, Chicago, Ill.*

Bigelow, Robert P., Ph.D., Assistant Professor of Zoölogy and Parasitology, *Massachusetts Institute of Technology, Boston, Mass.*

Black, Davidson, B.A., M.B., Assistant Professor of Anatomy, Western Reserve University, Medical Department, *1353 East 9th Street, Cleveland, Ohio.*

Blair, Vilray Papin, A.M., M.D., Clinical Professor of Surgery, Medical Department, Washington University, *Metropolitan Building, St. Louis, Mo.*

Blaisdell, Frank Ellsworth, M.D., Assistant Professor of Surgery, Medical Department of Stanford University, *1520 Lake Street, San Francisco, Calif.*

Blake, Joseph Augustus, A. B., M.D., *American Ambulance Hospital, Boulevard d'Inkermann, Neully-sur-Seine, Paris, France.*

Boyden, Edward Allen, Teaching fellow, Histology and Embryology, *Harvard Medical School, Boston, Mass.*

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Brödel, Max, Associate Professor of Art as Applied to Medicine, Johns Hopkins University, *Johns Hopkins Hospital, Baltimore, Md.*

Brooks, William Allen, A.M., M.D., *167 Beacon Street, Boston, Mass.*

Brown, A. J., M.D., Demonstrator of Anatomy, Columbia University, *156 East 64th Street, New York, N. Y.*

Browning, William, Ph.B., M.D., Professor of Nervous and Mental Diseases, Long Island College Hospital, *54 Lefferts Place, Brooklyn, N. Y.*

Bryce, Thomas H., M.A., M.D., Regius Professor of Anatomy, University of Glasgow, No. 2, *The University, Glasgow, Scotland.*

Bullard, H. Hays, A.M., Ph.D., Instructor in Anatomy and Neurology, *University of Pittsburgh Medical School, Pittsburgh, Pa.*

Bunting, Charles Henry, M.D., Professor of Pathology, *University of Wisconsin, 1804 Madison Street, Madison, Wis.*

Burrows, Montrose, T., A.B., M.D., Instructor in Anatomy, *Cornell University Medical College, New York, N. Y.*

Campbell, William Francis, A.B., M.D., Professor of Anatomy and Histology, Long Island College Hospital, *394 Clinton Avenue, Brooklyn, N. Y.*

Carpenter, Frederick Walton, Ph.D., Professor of Zoölogy, *Trinity College, Hartford, Conn.*

Chase, Martin Rist, M.S., Assistant in Anatomy, Northwestern University Medical School, *2431 Dearborn Street, Chicago, Ill.*

Cheever, David, M.D., Assistant Professor of Surgical Anatomy, Harvard Medical School, *355 Marlborough Street, Boston, Mass.*

Chidester, Floyd E., A.M., Ph.D., Assistant Professor of Zoölogy, *Rutgers College, New Brunswick, N. J.*

Chillingworth, Felix P., M.D., Assistant Professor of Physiology and Pharmcology, *Tulane University, New Orleans, La.*

Clapp, Cornelia Maria, Ph.D., Professor of Zoölogy, *Mount Holyoke College, South Hadley, Mass.*

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Clark, Eliot R., A.B., M.D., Professor of Anatomy, *University of Missouri, Columbia, Mo.*

Coghill, George E., Ph.D., Associate Professor of Anatomy, University of Kansas Medical School, *338 Illinois Street, Lawrence, Kansas.*

Cohn, Alfred E., M.D., Associate in Medicine, Rockefeller Institute for Medical Research, *315 Central Park West, New York, N. Y.*

Cohoe, Benson A., A.B., M.B., Associate Professor of Therapeutics, University of Pittsburgh, *705 North Highland Avenue, Pittsburgh, Pa.*

Conant, William Merritt, M.D., Instructor in Anatomy in Harvard Medical School, *486 Commonwealth Avenue, Boston, Mass.*

Conklin, Edwin Grant, A.M., Ph.D., Sc.D., Professor of Biology, Princeton University, *139 Broadmead Avenue, Princeton, N. J.*

Corner, George W., M.D., Assistant in Anatomy, Johns Hopkins University, *Johns Hopkins Medical School, Baltimore, Md.*

Corning, H. K., M.D., Professor of Anatomy, *Bündesstr. 17, Basel, Switzerland.*

Corson, Eugene Rollin, B.S., M.D., Surgeon, Lecturer on Anatomy, Savannah, Hospital Training School for Nurses, *10 Jones Street, West, Savannah, Ga.*

Cowdry, Edmund V., Ph.D., Associate in Anatomy, Anatomical Laboratory, *Johns Hopkins Medical School, Baltimore, Md.*

Craig, Joseph David, A.M., M.D., Professor of Anatomy, Albany Medical College, *12 Ten Broeck Street, Albany, N. Y.*

Crile, George W., M.D., Professor of Surgery, Western Reserve University, *1021 Prospect Avenue, Cleveland, O.*

Cullen, Thomas S., M.B., Associate Professor of Gynecology, Johns Hopkins University, *3 West Preston Street, Baltimore, Md.*

Cunningham, Robert S., B.S., A.M., Johns Hopkins Medical School, *716 N. Broadway, Baltimore, Md.*

Curtis, George M., A.B., A.M., Assistant Professor of Anatomy, Medical Department of the Vanderbilt University, *907 First Avenue, Nashville, Tenn.*

Dahlgren, Ulric, M.S., Professor of Biology, Princeton University, *204 Guyot Hall, Princeton, N. J.*

Danforth, Charles Haskell, A.M., Ph.D., Instructor in Anatomy, Medical Department, *Washington University, Medical School, St. Louis, Mo.*

Darrach, William, A.M., M.D., Assistant Attending Surgeon, Presbyterian Hospital, Instructor in Clinical Surgery, Columbia University, *47 West 50th Street, New York, N. Y.*

Davison, Alvin, M.A., Ph.D., Professor of Biology, *Lafayette College, Easton, Pa.*

Davis, David M., B.S., *Johns Hopkins Medical School, Baltimore, Md.*

Davis, Henry K., A.B., A.M., Instructor in Anatomy, *Cornell University Medical College, Ithaca, N. Y.*

Dawburn, Robert H. Mackay, M.D., Professor of Anatomy, New York Polyclinic Medical School and Hospital, *105 West 74th Street, New York, N. Y.*

Dean, Bashford, Ph.D., Professor of Vertebrate Zoölogy, Columbia University, Curator of Fishes and Reptiles, American Museum Natural History, *Riverdale-on-Hudson, New York.*

Dexter, Franklin, M.D., *247 Marlborough Street, Boston, Mass.*

Dixon, A. Francis, M.B., Sc.D., University Professor of Anatomy, Trinity College, *73 Grosvenor Road, Dublin, Ireland.*

Dodson, John Milton, A.M., M.D., Dean and Professor of Medicine, Rush Medical College, University of Chicago, *5806 Blackston Avenue, Chicago, Ill.*

Donaldson, Henry Herbert, Ph.D., D.Sc., (Ex. Com. '09–'13), Professor of Neurology, *The Wistar Institute of Anatomy and Biology, Woodland Avenue and 36th Street, Philadelphia, Pa.*

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Dunn, Elizabeth Hopkins, A.M., M.D., Nelson Morris Laboratory for Medical Research, *4760 Lake Park Avenue, Chicago, Ill.*

Eccles, Robert G., M.D., Phar.D., *681 Tenth Street, Brooklyn, N. Y.*

Edwards, Charles Lincoln, Ph.D., Director of Nature Study, Los Angeles City Schools, *1032 West 39th Place, Los Angeles, Calif.*

Eggerth, Arnold Henry, Assistant in Histology, *University of Michigan, Ann Harbor, Michigan.*

Elliot, Gilbert M., A.M., M.D., Demonstrator of Anatomy, Medical School of Maine, *152 Maine Street, Brunswick, Me.*

Emmel, Victor E., M.S., Ph.D., Associate Professor of Anatomy, Washington University Medical School, St. Louis, Mo.

Essick, Charles Rhein, B.A., M.D., Instructor in Anatomy, Johns Hopkins University, *1807 North Caroline Street, Baltimore, Md.*

Evans, Herbert McLean, B.S., M.D., Associate Professor of Anatomy, Research Associate in Embryology, Carnegie Institution, *Johns Hopkins Medical School, Baltimore, Md.*

Evatt, Evelyn John, B.S., M.B., Professor of Anatomy, *Royal College of Surgeons, Dublin, Ireland.*

Eycleshymer, Albert Chauncey, Ph.D., M.D., Professor of Anatomy, *Medical Department, University of Illinois, Honore and Congress Streets, Chicago, Ill.*

Ferris, Harry Burr, A.B., M.D., Hunt Professor of Anatomy and Head of the Department of Anatomy, Medical Department, Yale University, *395 St. Ronan Street, New Haven, Conn.*

Fetterolf, George, A.B., M.D., Sc.D., Assistant Professor of Anatomy, University of Pennsylvania, *330 South 16th Street, Philadelphia, Pa.*

Fischelis, Philip, M.D., Associate Professor of Histology and Embryology, Medico-Chirurgical College, *328 North 5th Street, Philadelphia, Pa.*

Flint, Joseph Marshall, B.S., A.M., M.D. (Second Vice-Pres. '00-'04), Professor of Surgery, Yale University, *320 Temple Street, New Haven, Conn.*

Frost, Gilman Dubois, A.M., M.D., Professor of Clinical Medicine, *Dartmouth Medical School, Hanover, N. H.*

Gage, Simon Henry, B.S. (Ex. Com. '06–'11), Emeritus Professor of Histology and Embryology, *Cornell University, Ithaca, N. Y.*

Gage, Mrs. Susanna Phelps, B.Ph., *4 South Avenue, Ithaca, N. Y.*

Gallaudet, Bern Budd, A.M., M.D., Assistant Professor of Anatomy, Columbia University, Consulting Surgeon Bellevue Hospital, *110 East 16th Street, New York, N. Y.*

Geddes, A. Campbell, M.D., M.B., Ch.B., F.R.S.E., Professor of Anatomy, *McGill University, Montreal, Canada.*

Gibson, James A., M.D., Professor of Anatomy, Medical Department, University of Buffalo, *24 High Street, Buffalo, N. Y.*

Gilman, Philip Kingsworth, B.A., M.D., Professor of Surgery, University of Philippines, *Suite 417–427 Kneedler Bldg., Manila, P. I.*

Goetsch, Emil, Ph.D., M.D., Associate in Surgery, *Harvard Medical School,* Resident Surgeon, *Peter Brigham Hospital, Boston, Mass.*

Greene, Charles W., Ph.D., Professor of Physiology and Pharmacology, *University of Missouri, Columbia, Mo.*

Greenman, Milton J., Ph.B., M.D., Sc.D., Director of the *Wistar Institute of Anatomy and Biology, 36th Street and Woodland Avenue, Philadelphia, Pa.*

GUDERNATSCH, J. F., Ph.D., Instructor in Anatomy, *Cornell University Medical College, New York City.*

GUILD, STACY R., A.M., Instructor in Histology and Embryology, University of Michigan, *1511 Washtenaw Avenue, Ann Arbor, Mich.*

GUYER, MICHAEL F., Ph.D., Professor of Zoölogy, University of Wisconsin, *138 Prospect Avenue, Madison, Wis.*

HALSTED, WILLIAM STEWART, M.D., Professor of Surgery, Johns Hopkins University, *1201 Eutaw Place, Baltimore, Md.*

HAMANN, CARL A., M.D., (Ex. Com. '02–'04), Professor of Applied Anatomy and Clinical Surgery, Western Reserve University, *416 Osborn Building, Cleveland, Ohio.*

HARDESTY, IRVING, A.B., Ph.D., (Ex. Com. '10 and '12–'15), Professor of Anatomy, *Tulane University of Louisiana, Station 20, New Orleans, La.*

HARE, EARL R., A.B., M.D., Instructor in Surgery, University of Minnesota, *623 Syndicate Building, Minneapolis, Minn.*

HARRISON, ROSS GRANVILLE, Ph.D., M.D. (Pres. '12–'13), Bronson Professor of Comparative Anatomy, *Yale University, New Haven, Conn.*

HARVEY, BASIL COLEMAN HYATT, A.B., M.B., Associate Professor of Anatomy, University of Chicago, *Department of Anatomy, University of Chicago, Chicago, Ill.*

HARVEY, RICHARD WARREN, M.S., M.D., Assistant Professor of Anatomy, Anatomy Department, University of California, *Berkeley, Calif.*

HATAI, SHINKISHI, Ph.D., Associate in Neurology, *Wistar Institute of Anatomy and Biology, Philadelphia, Pa.*

HATHAWAY, JOSEPH H., A.M., M.D., Professor of Anatomy, Anatomical Department, *Detroit Medical College, Detroit, Mich.*

HAZEN, CHARLES MORSE, A.M., M.D., Professor of Physiology, *Medical College of Virginia, Richmond, Bon Air, Va.*

HEISLER, JOHN C., M.D., Professor of Anatomy, Medico-Chirurgical College, *3829 Walnut Street, Philadelphia, Pa.*

HELDT, THOMAS JOHANES, A.B., A.M., *200 East Lanvale Street, Baltimore, Md.*

HERRICK, CHARLES JUDSON, Ph.D., (Ex. Com. 1913–) Professor of Neurology, University of Chicago, *Laboratory of Anatomy, University of Chicago, Chicago, Ill.*

HERTZLER, ARTHUR E., M.D., F.A.C.S., Associate in Surgery, University of Kansas, *1004 Rialto Building, Kansas City, Mo.*

HERZOG, MAXIMILIAN, M.D., Professor of Pathology and Bacteriology, La Gola University, *1358 Fulton Street, Chicago, Ill.*

HEUER, GEORGE JULIUS, B.S., M.D., Resident-Surgeon, Johns Hopkins Hospital, and Instructor in Surgery, *Johns Hopkins Hospital, Baltimore, Md.*

HEUSER, CHESTER H., A.M., Ph.D., Fellow in Anatomy, *Wistar Institute of Anatomy, 36th Street and Woodland Avenue, Philadelphia, Pa.*

HEWSON, ADDINELL, A.M., M.D., Professor of Anatomy, Philadelphia Polyclinic for Graduates in Medicine, *2120 Spruce Street, Philadelphia, Pa.*

HILL, HOWARD, M.D., *1010 Rialto Building, Kansas City, Mo.*

HILTON, WILLIAM A., Ph.D., Professor of Zoölogy, *Pomona College, Claremont, Calif.*

HODGE, C. F., Ph.D., Professor of Social Biology, *University of Oregon, Eugene, Oregon.*

HOEVE, HUBERTUS H. J., M.D., *Meherrin Hospital, Meherrin, Virginia.*

HOOKER, DAVENPORT, M.A., Ph.D., Assistant Professor of Anatomy, *University of Pittsburgh Medical School, Grant Boulevard, Pittsburgh, Pa.*

HOPKINS, GRANT SHERMAN, Sc.D., D.V.M., Professor of Veterinary Anatomy *Cornell University, Ithaca, N. Y.*

HOSKINS, ELMER R., A.B., A.M., Instructor in Anatomy, *University of Minnesota, Minneapolis, Minn.*

HRDLIČKA, ALES, M.D., Curator of the Division of Physical Anthropology, *United States National Museum, Washington, D. C.*

HUBER, G. CARL, M.D. (Second Vice-Pres. '00–'01, Secretary-Treasurer '02–'13, Pres. '14–) Professor of Anatomy and Director of the Anatomical Laboratories, University of Michigan, *1330 Hill Street, Ann Arbor, Mich.*

HUNTINGTON, GEORGE S., A.M., M.D., D.Sc., LL.D. (Ex. Com. '95–'97, '04–'07, Pres. '99–'03), Professor of Anatomy, Columbia University, *437 West 59th Street, New York, N. Y.*

INGALLS, N. WILLIAM, M.D., Associate Professor of Anatomy, *Medical College, Western Reserve University, Cleveland, Ohio.*

JACKSON, CLARENCE M., M.S., M.D., (Ex. Com. '10–'14), Professor and Head of the Department of Anatomy, *University of Minnesota, Institute of Anatomy, Minneapolis, Minn.*

JENKINS, GEORGE B., M.D., Professor of Anatomy, *Department of Anatomy, University of Louisville, Louisville, Ky.*

JOHNSON, CHARLES EUGENE, Ph.D., Instructor in Comparative Anatomy, Department of Animal Biology, *University of Minnesota, Minneapolis, Minn.*

JOHNSON, FRANKLIN P., A.M., Ph.D., Associate Professor of Anatomy, *University of Missouri, 408 South Ninth Street, Columbia, Mo.*

JOHNSTON, JOHN B., Ph.D., Professor of Comparative Neurology, University of Minnesota, *University of Minnesota, Minneapolis, Minn.*

JORDAN, HARVEY ERNEST, Ph.D., Professor of Histology and Embryology, *University of Virginia, University, Va.*

KAMPMEIER, OTTO FREDERICK, A.B., Ph.D., Instructor in Embryology and Anatomy, *University of Pittsburgh Medical School, Pittsburgh, Pa.*

KAPPERS, CORNELIUS, UBBO ARIENS, Director of the *Central Institute for Brain Research of Holland, Mauritskade 61, Amsterdam, Holland.*

KEILLER, WILLIAM, L.R.C.P. and F.R.C.S.Ed. (Second Vice-Pres. '98–'99), Professor of Anatomy, Medical Department University of Texas, *State Medical College, Galveston, Texas.*

KELLY, HOWARD ATWOOD, A.B., M.D., LL.D., Professor of Gynecology, Johns Hopkins University, *1418 Eutaw Place, Baltimore, Md.*

KERR, ABRAM T., B.S., M.D., (Ex. Com. '10–14), Professor of Anatomy, *Cornell University Medical College, Ithaca, N. Y.*

KINGSBURY, BENJAMIN F., Ph.D., M.D., Professor of Histology and Embryology, Cornell University, *802 University Avenue, Ithaca, N. Y.*

KINGSLEY, JOHN STERLING, Sc.D., Professor of Zoölogy, *University of Illinois, Urbana, Ill.*

KING, HELEN DEAN, A.B., Ph.D., Assistant Professor of Embryology, *Wistar Institute of Anatomy, 36th Street and Woodland Avenue, Philadelphia, Pa.*

KNOWER, HENRY McE., A.B., Ph.D., (Ex. Com. '11-15), Professor of Anatomy, *Medical Department, University of Cincinnati, Station V, Cincinnati, Ohio.*

KOFOID, CHARLES ATWOOD, Ph.D., Professor of Zoölogy University of California, Assistant Director San Diego Marine Biological Station, *2616 Etna Street, Berkeley, Calif.*

KUNKEL, BEVERLY WAUGH, Ph.B., Ph.D., Professor of Zoölogy, *Beloit College, Beloit, Wisconsin.*

KUNTZ, ALBERT, Ph.D., Department of Anatomy, University of St. Louis, *3911 Castleman Avenue, St. Louis, Mo.*

KUTCHIN, HARRIET LEHMANN, A.M., Assistant in Biology, University of Montana, *527 Ford Street, Missoula, Mont.*

KYES, PRESTON, A.M., M.D., Assistant Professor of Experimental Pathology, *Department of Pathology, University of Chicago, Chicago, Ill.*

LAMB, DANIEL SMITH, A.M., M.D., LL.D., (Secretary-Treasurer '90–'01, Vice-Pres. '02–'03) Pathologist Army Medical Museum, Professor of Anatomy, Howard University, Medical Department, *2114 18th Street N. W., Washington, D.C.*

LAMBERT, ADRIAN V.S., A.B., M.D., Associate Professor of Surgery, Columbia University, *168 East 71st Street, New York, N. Y.*

LANDACRE FRANCIS LEROY, A.B., Professor of Anatomy, Ohio State University, *2026 Inka Ave., Columbus, Ohio.*

LANE, MICHAEL ANDREW, B.S., *122 S. California Avenue, Chicago, Ill.*

LEE, THOMAS G., B.S., M.D. (Ex. Com. '08–'10, Vice Pres. '12–'13), Professor of Comparative Anatomy, University of Minnesota, *Institute of Anatomy, University of Minnesota, Minneapolis, Minn.*

LEIDY, JOSEPH, Jr., A.M., M.D., *1319 Locust Street, Philadelphia, Pa.*

LEWIS, DEAN D., M.D., Assistant Professor of Surgery, Rush Medical College, *People's Gas Building, Chicago, Ill.*

LEWIS, FREDERIC T., A.M., M.D., (Ex.Com. '09-'13, Vice-Pres. '14–), Assistant Professor of Embryology, *Harvard Medical School, Boston, Mass.*

LEWIS, WARREN HARMON, B.S., M.D., (Ex.Com. '09–'11, '14–), Professor of Physiological Anatomy, *Johns Hopkins University, Medical School, Baltimore, Md.*

LILLIE, FRANK RATHAY, Ph.D., Professor of Embryology, Chairman of Department of Zoölogy, University of Chicago; Director Marine Biological Laboratory, Woods Hole, Mass., *University of Chicago, Chicago, Ill.*

LINEBACK, PAUL EUGENE, A.B., M.D., Teaching Fellow in Histology and Embryology, *Harvard Medical School, Boston, Mass.*

LOCY, WILLIAM A., Ph.D., Sc.D., Professor of Zoölogy and Director of the Zoölogical Laboratory, Northwestern University, *1745 Orrington Avenue, Evanston, Ill.*

LOEB, HANAU WOLF, A.M., M.D., Professor and Director of the Department of the Diseases of the Ear, Nose and Throat, St. Louis University, *537 North Grand Avenue, St. Louis, Mo.*

LORD, FREDERIC P., A.B., M.D., Professor of Anatomy and Histology, *Dartmouth Medical School, Hanover, N. H.*

LOWREY, LAWSON GENTRY, A.M., *Harvard Medical School, Boston, Mass.*

MACKLIN, C. C., M.B., Assistant in Anatomy, *Johns Hopkins Medical School, Baltimore, Md.*

MCCARTHY, JOHN GEORGE, M.D., Formerly Assistant Professor of Anatomy, McGill University, *112 St. Mark Street, Montreal, Canada.*

McClure, Charles Freeman Williams, A.M., Sc.D. (Vice Pres. '10–'11, Ex. Com. '12–'16), Professor of Comparative Anatomy, *Princeton University. Princeton, N. J.*

McCormack, William Eli, M.D., Instructor in Embryology and Histology, *University of Louisville, Louisville, Ky.*

McCotter, Rollo E., M.D., Professor of Anatomy, *Medical Department, University of Michigan, Ann Arbor, Michigan.*

McFarland, Frank Mace, Ph.D., Professor of Histology, *Leland Stanford Junior University, Stanford, Calif.*

McGill, Caroline, A.M., Ph.D., M.D., Pathologist, *Murray Hospital, Butte, Montana.*

McKibben, Paul S., Ph.D., Professor of Anatomy, *Department of Anatomy, Western University, London, Canada.*

McMurrich, James Playfair, A.M., Ph.D., LL.D. (Ex. Com. '06–'07, Pres. '08–'09), Professor of Anatomy, University of Toronto, *75 Forest Hill Road, Toronto, Canada.*

McWhorter, John E., M.D., Worker under George Crocker Research Fund, College of Physicians and Surgeons, Columbia University, *205 West 107th Street, New York, N. Y.*

Mall, Franklin P., A.M., M.D., LL.D., D.Sc. (Ex.Com.'00–'05, Pres. '06–'07), Professor of Anatomy, *Johns Hopkins Medical School, Baltimore, Md.*

Mangum, Charles S., A.B., M.D., Professor of Anatomy, *University of North Carolina, Chapel Hill, N. C.*

Malone, Edward Fall, A.B., M.D., Assistant Professor of Anatomy, *University of Cincinnati, Station V, Cincinnati, O.*

Mark, Edward Laurens, Ph.D., LL.D., Hersey Professor of Anatomy and Director of the Zoölogical Laboratory, Harvard University, *109 Irving Street, Cambridge, Mass.*

Martin, Walton, Ph.B., M.D., Professor of Clinical Surgery, Columbia University, *25 West 50th Street, New York, N. Y.*

Matas, Rudolph, M.D., Professor of Surgery, Tulane University, *2255 St. Charles Avenue, New Orleans, La.*

Maximow, Alexander, M.D., Professor of Histology and Embryology at the Imperial Military Academy of Medicine, Petrograd, Russia, *Botkinskaja 2, Petrograd, Russia.*

Mellus, Edward Lindon, M.D., *10 Sewall Avenue, Brookline, Mass.*

Mercer, William F., Ph.D., Professor of Biology, Ohio University, *200 East State Street, Athens, Ohio.*

Metheny, D. Gregg, M.D., Associate in Anatomy, *Jefferson Medical College, 11th and Clinton Streets, Philadelphia, Pa.*

Meyer, Adolf, M.D., LL.D., Professor of Psychiatry and Director of the Henry Phipps Psychiatric Clinic, *Johns Hopkins Hospital, Baltimore, Md.*

Meyer, Arthur W., S.B., M.D., (Ex. Com. '12–'16), Professor of Human Anatomy, Leland Stanford Junior University, *Stanford University, Calif.*

Miller, Adam M., A.M., Professor of Anatomy, Long Island College Hospital, *Henry and Amity Streets, Brooklyn, N. Y.*

Miller, William Snow, M.D. (Vice-Pres. '08–'09), Associate Professor of Anatomy, University of Wisconsin, *415 West Wilson Street, Madison, Wis.*

MIXTER, SAMUEL JASON, B.S., M.D., Visiting Surgeon Massachusetts General Hospital, *180 Marlboro Street, Boston, Mass.*

MOODIE, ROY L., A.B., Ph.D., Assistant Professor of Anatomy, *University of Illinois Medical College, Chicago, Ill.*

MOODY, ROBERT ORTEN, B.S., M.D., Associate Professor of Anatomy, University of California, *2826 Garber Street, Berkeley, Calif.*

MORGAN, JAMES DUDLEY, A.B., M.D., Physician, Garfield Hospital, *919 15th Street, McPherson Square, Washington, D. C.*

MORRILL, CHARLES V., Ph.D, Instructor in Anatomy, New York University and Bellevue Medical College, *338 East 26th Street, New York, N. Y.*

MULLER, HENRY R., M.D., Assistant in Anatomy, *Johns Hopkins Medical School, Baltimore, Md.*

MUNSON, JOHN P., Ph.D., Head of the Department of Biology, Washington State Normal School, *706 North Anderson Street, Ellensburg, Washington.*

MURPHEY, HOWARD S., D.V.M., Professor of Anatomy and Histology, Ames, Ia. *519 Welch Avenue, Station A., Ames, Ia.*

MYERS, BURTON D., A.M., M.D., Professor of Anatomy and Secretary of the Indiana University School of Medicine, *Indiana University, Bloomington, Ind.*

MYERS, JAY A., A.B., Ph.D., Instructor in Anatomy, *University of Minnesota, Minneapolis, Minn.*

NACHTRIEB, HENRY FRANCIS, B.S., Professor of Animal Biology and Head of the Department, University of Minnesota, *905 East 6th Street, Minneapolis, Minn.*

NEAL, HERBERT VINCENT, Ph.D., Professor of Zoölogy, Tufts College, *Tufts College, Mass.*

NEWMAN, HORATIO HACKETT, Ph.D., Associate Professor of Zoölogy, University of Chicago, *Department of Zoölogy, University of Chicago, Chicago, Ill.*

NOBLE, HARRIET ISABEL, *262 Putnam Avenue, Brooklyn, N. Y.*

NORRIS, H. W., B.S., A.M., Professor of Zoölogy, *Grinnell College, Grinnell, Iowa.*

PAPEZ, JAMES WENCESLAS, B.A., M.D., Professor of Anatomy, *Atlanta Medical College, Atlanta, Ga.*

PARKER, GEORGE HOWARD, D.Sc., Professor of Zoölogy, Harvard University, *16 Berkeley Street, Cambridge, Mass.*

PATON, STEWART, A.B., M.D., Lecturer in Biology, *Princeton University, Princeton, N. J.*

PATTEN, WILLIAM, Ph.D., Professor of Zoölogy, *Dartmouth College, Hanover, N.H.*

PATERSON, A. MELVILLE, M.D., F.R.C.S., Professor of Anatomy, *University of Liverpool, Liverpool, England.*

PATTERSON, JOHN THOMAS, Ph.D., Professor and Chairman of the School of Zoölogy, University of Texas, *University Station, Austin, Texas.*

PIERSOL, GEORGE A., M.D., Sc.D. (Vice-Pres. '93–'94, '98–'99, '06–'07, Pres. '10–'11) Professor of Anatomy, University of Pennsylvania, *4724 Chester Avenue, Philadelphia, Pa.*

PIERSOL, WILLIAM HUNTER, A.B., M.B., Associate Professor of Histology and Embryology, *Biological Department University of Toronto, Toronto, Canada.*

POHLMAN, AUGUSTUS G., M.D., Professor of Anatomy, *Medical Department, University of St. Louis, St. Louis, Mo.*

POTTER, PETER, M.S., M.D., Oculist and Aurist, Murray Hospital, Butte, Montana, *411–413 Hennessy Building, Butte, Montana.*

PRENTISS, CHARLES W., A.M., Ph.D., Professor of Microscopic Anatomy, *Northwestern University Medical School, 2421 Dearborn Street, Chicago, Ill.*

PRENTISS, H. J., M.D., M.E., Professor of Anatomy, University of Iowa, *Iowa City, Iowa.*

PRYOR, JOSEPH WILLIAM, M.D., Professor of Anatomy and Physiology, State College of Kentucky, *261 North Broadway, Lexington, Ky.*

RADASCH, HENRY E., M.S., M.D., Assistant Professor of Histology and Embryology, Jefferson Medical College, *Daniel Baugh Institute of Anatomy, 11th and Clinton Streets, Philadelphia, Pa.*

RANSON, STEPHEN W., M.D., Ph.D., Professor of Anatomy, Northwestern University Medical School, *2431 Dearborn Street, Chicago, Ill.*

REAGAN, FRANKLIN P., A.B., *Princeton University, Princeton, N. J.*

REED, HUGH DANIEL, Ph.D., Assistant Professor of Zoölogy, Cornell University, *108 Brandon Place, Ithaca, N. Y.*

REESE, ALBERT MOORE, A.B., Ph.D., Professor of Zoölogy, *West Virginia University, Morgantown, W. Va.*

RETZER, ROBERT, M.D., Assistant Professor of Anatomy, University of Chicago, *Department of Anatomy, University of Chicago, Chicago, Ill.*

REVELL, DANIEL GRAISBERRY, A.B., M.B., Provincial Pathologist, Bacteriologist and Analyst of the Provincial Laboratory, *901 Eighth Street, N. W., Strathcona, Alberta, Canada.*

RHINEHART, D.A., M.D., Professor of Anatomy, University of Arkansas, *Old State House, Little Rock, Arkansas.*

RICE, EDWARD LORANUS, Ph.D., Professor of Zoölogy, *Ohio Wesleyan University, Delaware, Ohio.*

ROBINSON, ARTHUR, M.D., F.R.C.S. (Edinburgh) Professor of Anatomy, University of Edinburgh, *The University, Edinburgh, Scotland.*

RUTH, EDWARD S., M.D., Professor of Anatomy, Southern Methodist University Medical Department, *4123 Bryan Street, Dallas, Texas.*

SABIN, FLORENCE R., B.S., M.D., (Second Vice-Pres. '08–'09), Associate Professor of Anatomy, *Johns Hopkins University, Medical Department, Baltimore, Md.*

SACHS, ERNEST, A.B., M.D., Associate in Surgery, Washington University Medical School, *St. Louis, Mo.*

SAMPSON, JOHN ALBERTSON, A.B., M.D., Professor of Gynecology, Albany Medical College, *180 Washington Avenue, Albany, N. Y.*

SANTEE, HARRIS E., Ph.D., M.D., Professor of Anatomy, Jenner Medical College, and Professor of Neural Anatomy, Chicago College of Medicine and Surgery, *2806 Warren Avenue, Chicago, Ill.*

SCAMMON, RICHARD E., Ph.D., Associate Professor of Anatomy, *Institute of Anatomy, University of Minnesota, Minneapolis, Minn.*

SCHAEFER, MARIE CHARLOTTE, M.D., Associate Professor of Biology, Histology and Embryology, *Medical Department, University of Texas, Galveston, Texas.*

SCHAEFFER, JACOB PARSONS, A.M., M.D., Ph.D., Professor of Anatomy, *Jefferson Medical College, 11th and Clinton Streets, Philadelphia, Pa.*

SCHOEMAKER, DANIEL M., B.S., M.D., Professor of Anatomy, Medical Department, St. Louis University, *1402 South Grand Avenue, St. Louis, Mo.*

SCHULTE, HERMANN VON W., A.B., M.D., (Ex.Com. '15–) Associate Professor of Anatomy, Columbia University, *206 West 86th Street, New York, N. Y.*

SCHMITTER, FERDINAND, A.B., M.D., *Captain Medical Corps, U. S. Army, Department Hospital, Manila, P. I.*

SCOTT, KATHERINE JULIA, A.B., *Johns Hopkins Medical School, Baltimore, Md.*

SEELIG, MAJOR G., A.B., M.D., Professor of Surgery, St. Louis University, *Humboldt Building, 537 North Grand Avenue. St. Louis, Mo.*

SELLING, LAWRENCE, A.B., M.D., *789 Lovejoy Street, Portland, Oregon.*

SENIOR, HAROLD D., M.B., F.R.C.S., D.Sc., Professor of Anatomy, New York University, University and Bellevue Hospital Medical College, *338 East 26th Street, New York, N. Y.*

SHELDON, RALPH EDWARD, A.M., M.S., Ph.D., Associate Professor of Anatomy, *University of Pittsburgh Medical School, Grant Boulevard, Pittsburgh, Pa.*

SHIELDS, RANDOLPH TUCKER, A.B., M.D., Dean, *University of Nanking Medical School, Nanking, China.*

SHIPLEY, PAUL G., M.D., Assistant in Anatomy, John Hopkins University, *Johns Hopkins Medical School, Baltimore, Md.*

SHUFELDT, R. W., M.D., Major Medical Corps, U. S. A. (Retired)., *3356 Eighteenth Street, N. W., Washington, D. C.*

SILVESTER, CHARLES FREDERICK, Curator of the Zoölogical Museum and Assistant in Anatomy, Princeton University, *10 Nassau Hall, Princeton, N. J.*

SIMPSON, SUTHERLAND, M.D., D.Sc., F.R.S.E. (Edin.), Professor of Physiology, *Cornell University, Ithaca, N. Y.*

SISSON, SEPTIMUS, B.S., V.S., Professor of Comparative Anatomy, Ohio State University, *318 West 9th Avenue, Columbus, Ohio.*

SLUDER, GREENFIELD, M.D., Clinical Professor of Diseases of the Nose and Throat, Washington University Medical School, *3542 Washington Avenue, St. Louis, Mo.*

SMITH, CHARLES DENNISON, A.M., M.D., Superintendent Maine General Hospital, Professor of Physiology, Medical School of Maine, *Maine General Hospital, Portland, Me.*

SMITH, GEORGE MILTON, A.B., M.D., Associate in Pathology, Washington University Medical School, *St. Louis, Mo.*

SMITH, GRAFTON ELLIOT, M.A., M.D., F.R.S., Professor of Anatomy, *University of Manchester, 4 Willow Bank, Fallowfield, Manchester, England.*

SMITH, J. HOLMES, M.D., Professor of Anatomy, University of Maryland, *Green and Lombard Streets, Baltimore, Md.*

SMITH, PHILIP EDWARD, M.S., *Department of Anatomy, University of California, Berkeley, Calif.*

SNOW, PERRY G., A.B., Professor of Anatomy, School of Medicine, *University of Utah, Salt Lake City, Utah.*

SPITZKA, EDWARD ANTHONY, M.D., *66 East 73d Street, New York, N. Y.*

STEENSLAND, HALBERT SEVERIN, B.S., M.D., Professor of Pathology and Bacteriology, and Director of the Pathological Laboratory, College of Medicine, Syracuse University, *309 Orange Street, Syracuse, N. Y.*

STILES, HENRY WILSON, M.D., Professor of Anatomy, *College of Medicine, Syracuse University, 309 Orange Street, Syracuse, N. Y.*

STOCKARD, CHARLES RUPERT, M.S., Ph.D., (Secretary-Treasurer '14–) Professor of Anatomy, *Cornell University Medical College, New York, N.Y.*

STOTSENBURG, JAMES M., M.D., Associate in Anatomy, *Wistar Institute of Anatomy and Biology, Philadelphia, Pa.*

STREETER, GEORGE L., A.M., M.D., Research Associate in Embryology, Carnegie Institution, *Johns Hopkins Medical School, Baltimore, Md.*

STROMSTEN, FRANK ALBERT, D.Sc., Assistant Professor of Animal Biology, University of Iowa, *943 Iowa Avenue, Iowa City, Iowa.*

STRONG, OLIVER S., A.M., Ph.D., Instructor in Anatomy, Columbia University, *437 West 59th Street, New York, N. Y.*

STRONG. REUBEN MYRON, Ph.D., Professor of Anatomy, *University of Mississippi, Oxford, Mississippi.*

SUNDWALL, JOHN, Ph.D., Professor of Anatomy, *University of Kansas, Lawrence, Kansas.*

SYMINGTON, JOHNSON, M.D., F.R.S., Professor of Anatomy, *Queens University, Belfast, Ireland.*

SWIFT, CHARLES H., M.D., Ph.D., Associate in Anatomy, *Department of Anatomy, University of Chicago, Chicago, Ill.*

TAUSSIG, FREDERICK JOSEPH, A.B., M.D., Lecturer in Gynecology, Washington University Medical School, *4506 Maryland Avenue, St. Louis, Mo.*

TAYLOR, EDWARD W., A.M., M.D., Instructor in Neurology, Harvard Medical School, *457 Marlboro Street, Boston, Mass.*

TERRY, ROBERT JAMES, A.B., M.D. (Ex.Com. '08–'12), Professor of Anatomy, *Washington University Medical School, St. Louis, Mo.*

THOMPSON, ARTHUR, M.A., M.B., F.R.C.S., Professor of Anatomy, *University of Oxford, Department of Human Anatomy, Museum, Oxford, England.*

THORKELSON, JACOB, M.D., Professor of Anatomy, *College of Physicians and Surgeons, Baltimore, Md.*

THRO, WILLIAM C., A.M., M.D., Assistant Professor of Clinical Pathology, *Cornell University Medical College, 28th Street and 1st Avenue, New York, N. Y.*

THYNG, FREDERICK WILBUR, Ph.D., Assistant Professor of Anatomy in the *University and Bellevue Hospital Medical College, 26th Street and 1st Avenue, New York, N. Y.*

TILNEY, FREDERICK, A.B., M.D., Assistant Professor of Neurology, Columbia University, *161 Henry Street, Brooklyn, N. Y.*

TOBIE, WALTER E., M.D., Professor of Anatomy, Medical School of Maine, *3 Deering Street, Portland, Me.*

TODD, THOMAS WINGATE, M.B., Ch.B. (Manc.), F.R.C.S. (Eng.) Professor of Anatomy, *Medical Department, Western Reserve University, Cleveland, O.*

TRACY, HENRY C., A.M., Ph.D., Professor of Anatomy, Marquette University School of Medicine, *Fourth and Reservoir Street, Milwaukee, Wis.*

TUCKERMAN, FREDERICK, M.D., Ph.D., *16 College Street, Amherst, Mass.*

TUPPER, PAUL YOER, M.D., Clinical Professor of Surgery, Washington University Medical School, *Wall Building, St. Louis, Mo.*

WAITE, FREDERICK CLAYTON, A.M., Ph.D., Professor of Histology and Embryology, Western Reserve University, *1353 East 9th Street, Cleveland, Ohio.*

WALKER, GEORGE, M.D., Instructor in Surgery, Johns Hopkins University, *corner Charles and Centre Streets, Baltimore, Md.*

WALLIN, IVAN E., B.S., M.A., Assistant in Anatomy, Cornell University Medical College, *28th Street and First Avenue, New York, N. Y.*

WARREN, JOHN, M.D., Assistant Professor of Anatomy, *Harvard Medical School, Boston, Mass.*

WATERSTON, DAVID, M.A., M.D., F.R.C.S.Ed., Professor of Anatomy, *University of London, Kings College, Harlaw, Northwood, Middlesex, England.*

WEED, LEWIS HILL, A.M., M.D., Instructor in Anatomy, *Johns Hopkins Medical School, Baltimore, Md.*

WEIDENREICH FRANZ, M.D., a.o. Professor and Prosector of Anatomy, *19 Vogesen Street, Strassburg, i. Els. Germany.*

WEISSE, FANEUIL D., M.D. (Second Vice-Pres. '88–'89), Professor of Anatomy, New York College of Dentistry, *108 East 30th Street, New York, N. Y.*

WERBER, ERNEST I., Ph.D., *Department of Biology, Princeton University, Princeton, N. J.*

WEST, CHARLES IGNATIUS, M.D., Associate Professor of Anatomy, Medical Department of Howard University, *924 M Street N. W., Washington, D. C.*

WEST, P. A., B.A., *Johns Hopkins Medical School, Baltimore, Md.*

WEST, RANDOLPH, A.M., Student, College of Physicians and Surgeons, Columbia University, *24 West 59th Street, New York, N. Y.*

WEYSSE, ARTHUR WISSLAND, A.M., M.D., Ph.D., Professor of Biology and of Experimental Physiology, Boston University, *688 Boylston Street, Boston, Mass.*

WHIPPLE, ALLEN O., B.S., M.D., Instructor in Clinical Surgery, Columbia University, *981 Madison Avenue, New York, N. Y.*

WHITE, HARRY OSCAR, M.D., Professor of Anatomy, Histology and Embryology, *Medical Department, University of Southern California, Los Angeles, Calif.*

WHITEHEAD, RICHARD HENRY, A.B., M.D., LL.D., Professor of Anatomy, *University of Virginia, University P.O., Va.*

WIEMAN, HARRY LEWIS, Ph.D., Assistant Professor of Zoölogy *University of Cincinnati, Cincinnati, Ohio.*

WILDER, HARRIS HAWTHORNE, Ph.D., Professor of Zoölogy, Smith College, *Plymouth Inn, Northampton, Mass.*

WILLIAMS, STEPHEN RIGGS, Ph.D., Professor of Zoölogy, Miami University, *300 East Church Street, Oxford, O.*

WILLARD, WILLIAM A., A.M., Ph.D., Professor of Histology and Embryology, *University of Nebraska, College of Medicine, 42d Street and Dewey Avenue, Omaha, Nebraska.*

WILSON, J. GORDEN, M.A., M.B., C.M. (Edin.), Professor of Otology, Northwestern University Medical School, *2437 Dearborn Street, Chicago, Ill.*

WILSON, JAMES THOMAS, M.B., F.R.S., Challis Professor of Anatomy, *University of Sydney, Australia.*

WILSON, LOUIS BLANCHARD, M.D., Director of Laboratories, Mayo Clinic, *830 West College Street, Rochester, Minn.*

WINSLOW, GUY MONROE, Ph.D., Instructor in Histology, Tufts Medical College, *145 Woodland Road, Auburndale, Mass.*

WITHERSPOON, THOMAS CASEY, M.D., *307 Granite Street, Butte, Montana.*

WORCESTER, JOHN LOCKE, M.D., Instructor in Anatomy, University of Michigan, *1214 Willard, Ann Arbor, Michigan.*